J Clin Periodontol 2002: 29: 604–614
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Journal of Clinical Periodontology
0303-6979

Relationship of changes in interleukin-8 levels and granulocyte elastase activity in gingival crevicular fluid to subgingival periodontopathogens following non-surgical periodontal therapy in subjects with chronic periodontitis

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Jin LJ, Leung WK, Corbet EF, Söder B: Relationship of changes in interleukin-8 levels and granulocyte elastase activity in gingival crevicular fluid to subgingival periodontopathogens following non-surgical periodontal therapy in subjects with chronic periodontitis. J Clin Periodontol 2002; 29: 604–614. © Blackwell Munksgaard, 2002.

Abstract
Objectives: To determine the effect of scaling and root planing (SRP) on the interrelations of subgingival periodontopathogens and both interleukin-8 (IL-8) and granulocyte elastase activity in gingival crevicular fluid (GCF), and to assess their relations to the short-term treatment response in management of chronic periodontitis.
Material and methods: GCF and subgingival plaque were collected from 16 subjects with untreated chronic periodontitis at baseline and 4 weeks after SRP. IL-8 levels were determined by ELISA. Granulocyte elastase activity was analyzed with a specific substrate, pGluProVal-pNA, and the maximal rate of elastase activity (MR-EA) was calculated. 5 DNA-probes were used to detect the presence of *A. actinomycetemcomitans* (*A. a.*), *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*), and *T. denticola* (*T.d.*), with a sensitivity = 10^3 cells/paper point.
Results: IL-8 and MR-EA levels in GCF decreased significantly after SRP ($p < 0.001$) with a corresponding reduction of total count of the species. Of the sites with probing depth (PD) ≥ 5.0 mm and co-infection by *B.f., P.g., P.i. & T.d.* at baseline, the sites without persistent co-infection of these species after SRP exhibited a significant reduction of IL-8 levels ($p < 0.02$), MR-EA levels ($p < 0.02$) and PD ($p < 0.01$). No such change was found in the sites where such a co-infection persisted. Moreover, reduction of IL-8 levels in those pocket sites was accompanied by a concomitant reduction of MR-EA ($p < 0.02$) and PD ($p < 0.01$), while no significant change in MR-EA levels and PD was noted in those pocket sites that exhibited an increase of IL-8 levels after SRP. At baseline, the former group of sites showed significantly higher IL-8 levels than the latter group of sites ($p < 0.02$).
Conclusions: IL-8-related granulocyte elastase activity was related to the change in infection patterns of the target periodontopathogens following scaling and root planing. Varying initial IL-8 levels in GCF and a corresponding shifting change of granulocyte elastase activity in GCF may characterize the different short-term treatment responses.

Key words: chronic periodontitis; gingival crevicular fluid; granulocyte elastase; interleukin-8; periodontopathogen

Accepted for publication 16 July 2001

The current understanding of the pathogenesis of periodontitis suggests that the modulation of host response by bacterial products, such as lipopolysaccharide (LPS), is an important determinant of the onset and progression of periodontal diseases, which may stimulate the inflammatory process by up-regulating the host cells in the release of series inflammatory cytokines, mediators and enzymes capable of destroying host tissues (Van Dyke et al. 1993) and/or inhibits the local host defence function (Darveau et al. 1997, Madianos et al. 1997). Neutrophilic granulocytes play a pivotal role in the maintenance of host–bacteria homeostasis in periodontitis (Jin et al. 2000a). They migrate to inflamed gingival tissue in much higher numbers than other host defence cells and predominate in the gingival pocket epithelium and in the adjacent connective tissue (Christersson et al. 1987). Effective host response to bacterial challenge is primarily mediated by neutrophils and is characterized by an influx of neutrophils into gingival crevice. It has been suggested that the evasion of neutrophil clearance is a requisite for disease acquisition and the individual host response plays a primary role in determining the severity of disease expression (Offenbacher et al. 1993). A potentially destructive role has been proposed during the interplay of neutrophilic granulocytes with micro-organisms (Wilton 1986) due to high content of proteinases and generation of superoxide and reactive oxygen derivatives. The role of granulocytes in the pathogenesis of periodontal diseases has been studied extensively. An imbalance among extracellular release of proteinase and free oxygen radicals from granulocytes and proteinase inhibitors has been proposed as one of the major mechanisms of tissue destruction (Wewers et al. 1988).

Neutrophilic granulocyte recruitment and influx into gingival crevice depend upon chemotactic agonists that are derived from infecting micro-organisms and/or from the host, such as peptides of the N-formyl-methionyl-leucyl-phenylalanine type (FMLP) and other N-formylmethionyl peptides, C5a, leukotriene B_4 (LTB_4) and the alkylphospholipid derivative platelet activating factor (PAF) (Bickel 1993). In recent years, considerable interest has been focussed on the critical role of cytokines in the pathogenesis of periodontal destruction, not only as important mediators of host defence against the microbial challenge but as mediators of periodontal tissue destruction (Gemmell et al. 1997).

Interleukin-8 (IL-8) is a potent chemokine with a distinct target for recruitment and activation of human granulocytes and mediation of inflammatory processes (Baggiolini et al. 1989). It can be secreted from many different host cells, including monocytes/macrophages, lymphocytes, fibroblasts, endothelial cells and epithelial cells (Baggiolini et al. 1989, Lawlor et al. 1992). Recent studies have indicated that neutrophils *per se* have the ability to produce IL-8 in response to LPS from periodontopathogens (Yoshimura et al. 1997). Other than bacterial endotoxin, the main inducers of IL-8 secretion are IL-1, TNF-α and immune complexes (Schluger & Rom 1997). IL-8 plays an important role in regulation of neutrophil function (Tonetti 1997). It exerts various effects on activity and function of neutrophils, including inducing their adhesion to endothelial cells, shape change and transmigration, chemotaxis, exocytosis of primary and secondary granules containing series of lysosomal enzymes, and the respiratory burst. IL-8 has been shown to be highly important in the initiation and development of inflammatory response and tissue destruction in periodontal diseases, due to its critical role in recruitment and functional activation of neutrophilic granulocytes. By its nature, IL-8 is of considerable interest for a better understanding of the mechanisms leading to neutrophil-related tissue destruction (Bickel 1993).

Elastase (EC 3.4.21.37; 28 kDa) is a neutral serine proteinase stored in the cytoplasmic azurophil granules of granulocytes in amounts ranging up to 3 pg per cell. It participates intracellularly in phagocytosis, but it can also be released extracellularly by triggered granulocytes together with free oxygen radicals. Elastase is by far the highest of any proteinase quantitated in gingival crevicular fluid (GCF) during periodontal inflammation (Cox & Eley 1989). It has also been suggested to be a potential indicator of periodontal disease and disease progression (Palcanis et al. 1992, Armitage et al. 1994). Our earlier study has shown that GCF granulocyte elastase activity clearly correlated with long-term periodontal treatment response (Jin et al. 1995a). Significantly increased elastase levels were observed in subjects with refractory periodontitis, compared to the subjects who had a similar degree of periodontitis at baseline but who exhibited favourable treatment responses (Jin et al. 1995b). Assessment of granulocyte elastase activity in GCF provides a marker of intracrevicular granulocyte activity (Lamster 1997).

There is little information on the association of IL-8 with granulocyte elastase activity in response to the bacterial challenge in periodontal disease (Jin et al. 2000a), and their relation to treatment response and disease progression. Our recent study showed that there is an inverse relationship between IL-8 concentrations and the granulocyte elastase activity in GCF and that the local host response to the bacterial challenge in untreated periodontal pockets is diverse in terms of the intensity of inflammatory response measured by IL-8 associated granulocyte elastase activity in GCF (Jin et al. 1999, 2000a). In this study, we further examined (i) the effect of scaling and root planing (SRP) on the interrelations of subgingival periodontopathogens and both IL-8 and granulocyte elastase activity in GCF, and (ii) their relations to the short-term treatment response among a group of chronic periodontitis patients who had never received any prior periodontal therapy.

Material and methods

Patients and site selection

Sixteen Chinese adults, aged 32–55 years, were recruited for the study, as described in our previous study (Jin et al. 2000a). In brief, each subject had at least 20 natural teeth excluding 3rd molars. All subjects had untreated advanced chronic periodontitis, with $\geqslant$5.0 mm of probing depth, $\geqslant$3.0 mm of clinical attachment loss and radiographic evidence of alveolar bone loss on at least two teeth per quadrant. Their general health was good and none had received antibiotics within the preceding 6 months. None reported receiving any prior periodontal treatment or immunosuppressive therapy. Written and oral informed consent was obtained from all recruits and the study protocol was approved by the Ethical Committee, Faculty of Dentistry, the University of Hong Kong.

All patients were screened for their suitability and selection of sampling sites. The following parameters were measured at six sites on each tooth:

plaque index (PI) (Silness & Löe 1964), gingival index (GI) (Löe & Silness 1963), bleeding on probing (BOP), probing pocket depth (PD) and clinical attachment level (CAL). PD and CAL were measured using an electronic probe system (pocket probe and disk probe, Florida Probe® Co., Gainesville, FL, USA) by a calibrated examiner, and recorded in an interval scale, precise to 0.2 mm (Jin et al. 2000a,b). BOP was recorded as present or absent within 30 s after probing with the Florida Probe (pocket probe). Periapical radiographs were taken using a standardized long-cone paralleling technique. Five to seven nonadjacent sites, at least one per quadrant, were selected to be the sampling sites in each patient. The sites were classified according to the degree of gingival inflammation, PD, BOP and attachment loss (AL) into three categories (Jin et al. 2000a):

1 clinically healthy: GI $\leqslant$ 1, BOP (–), PD $\leqslant$ 3.0 mm, AL $\leqslant$ 1.0 mm, and bone loss (–);
2 gingivitis: GI $\geqslant$ 2, BOP (+), PD $\leqslant$ 4.0 mm, AL $\leqslant$ l.0 mm, and bone loss (–);
3 periodontitis: PD $\geqslant$ 5.0 mm, BOP (+), AL $\geqslant$ 3.0 mm, and bone loss (+).

When available, three to four periodontitis sites with the deepest PD, one to two gingivitis sites and one clinically healthy site were selected in each subject for sampling.

Collection of GCF

Prior to GCF collection, the supragingival plaque (PI) was scored. The sites to be sampled were isolated with cotton rolls, air-dried gently and supragingival plaque was carefully removed. A standard filter strip (Periopaper® GCF strips, IDE Interstate, Amityville, NY, USA) was inserted into the gingival crevice or pocket until mild resistance was felt and left in place for 30 s (Jin et al. 1999, 2000a). GCF volume was immediately determined using a GCF meter (Periotron 6000®, IDE Interstate). The readings from the Periotron 6000 were then converted to an actual volume (μL) by reference to a standard curve relating digital unit to volume, which was constructed using standard human serum. The electrode jaws were cleaned and dried, and the digital display was re-set to zero after each measurement. The strips were placed into 200 μL of sterile phosphate buffered saline (PBS, pH 7.2) for 1 h with constant agitation to elute the GCF sample. The eluate was aliquoted and then stored at −70 °C until analysed. Samples visually contaminated with blood were discarded. PD, BOP, GI, CAL and AL were recorded after sampling to confirm the periodontal status of the sites sampled.

Enzyme linked immunosorbent assay (ELISA) for human IL-8

The Quantikine human IL-8 ELISA kit (R & D Systems, Minneapolis, MN, USA) was used to quantitate the level of IL-8 in the GCF supernatants according to the manufacturer's instructions. The levels of IL-8 were determined as total amount per site (pg/site) and as concentration by dividing the amount of IL-8 by the GCF volume (pg/μL).

Assay of granulocyte elastase activity

Elastase activity was measured with a low molecular weight, chromogenic substrate specific for granulocyte elastase, L-pyroglutamyl-L-prolyl-L-valine-p-nitroanilide (pGluProVal-pNA, Kramps et al. 1983) (S-2484, AB Kabi Diagnostica, Stockholm, Sweden) according to our established method (Jin et al. 1999, 2000a). In brief, to 50 μL of sample, 34 μL of substrate working solution was added. The absorbance at 405 nm was measured immediately (0 h) in a Microplate Reader (Model 3550, Bio-Rad Laboratories, Hercules, CA, USA). Following the baseline measurement, the microplate was sealed and incubated at 37 °C, and measurements were taken at 15 min, 30 min, 45 min, 1 h, 2 h, 3 h, 4 h and 5 h. The rate of chromogen release represents the dynamics of elastase activity. All subsequent readings were subtracted by the 0 h-value (△O.D. at 405 nm). For each sample, the dynamic activity of elastase was plotted against time with a graphic software (StatView 512 + ™, BrainPower, Inc., Calabasas, CA, USA). The highest slope portion of a plotted substrate hydrolysis time course within the 5-h experimental period was used to calculate the maximal rate of elastase activity (MR-EA), expressed as mAbs/min per site.

Microbial sampling and detection

Following the GCF sampling, the subgingival plaque samples were collected from the same investigation sites by using a paper point (Jin et al. 1999, 2000a). In brief, a sterile, medium-sized absorbent paper point (Johnson & Johnson, New Brunswick, NJ, USA), was gently inserted into the depth of the gingival crevice/pocket and left in place for 10 s. The specimen-laden paper point was kept dry and placed in a capped vial. The samples were processed and analysed by an established method (DMDx-Patho Tek, ANAWA Laboratories, Zürich, Switzerland). Five species-specific DNA probes were used to detect the presence of *Actinobacillus actinomycetemcomitans* (*ATCC 43718*), *Bacteroides forsythus* (*ATCC 43037*), *Porphyromonas gingivalis (ATCC 33277), Prevotella intermedia* (*ATCC 33563*), and *Treponema denticola* (*ATCC 35405*) in a single sample. This assay has a minimum sensitivity of 10^3 cells/sample and a maximum sensitivity of 10^5 cells/sample. The results for individual samples were presented quantitatively within the detectable levels. For computation and statistical purposes, sites reported as $\geqslant 10^5$ cells/sample were considered to have a value of 10^5 cells.

Periodontal therapy

Following sampling, the patients received a course (total 4–5 h) of basic periodontal therapy consisting of oral hygiene instruction, scaling and root debridement as described previously (Jin et al. 2000b). Oral hygiene was reenforced at every visit during the 6-week treatment period. All the treatment was performed by a single senior staff hygienist. All subjects returned 4 weeks later for collection of GCF and subgingival plaque samples as described above. Clinical re-examination was performed according to the protocols at baseline.

Statistical analysis

The subjects were regarded as the main unit of observation and the individual sites as dependent subunits of observation. Presence of bacteria, singly or in combination, was expressed as the number and percentage of subjects and sites with detectable levels of bacteria. Before the analysis, natural logarithm

transformations, $X' = \log(X+1)$, of IL-8, elastase and bacterial data were used to generate distributions that more closely resembled the normal distribution and to stabilize the variance. These values under healthy and diseased conditions were calculated as subject averages, respectively.

The change from baseline to 1 month post-treatment was calculated for each parameter. Values for the variables were averaged within a subject and then averaged across individuals for each pre- and post-therapy visit. Significance of differences in mean levels of bacterial species was determined using the non-parametric Wilcoxon paired signed-rank tests. Multiple comparisons were made to assess the significance of differences in quantitative variables between the groups both at baseline and after the treatment as well as pre- and post-treatment difference within the group. A Bonferroni/Dunn correction was applied and a statistical significance level was adjusted accordingly. The differences between data sets with a probability of less than $0.05/\sqrt{n}$ were regarded as statistically significant, where n refers to the number of comparisons made (Brown & Beck 1988). The data analysis was performed using the StatView® 4.51 statistical package (Abacus Concepts, Inc. Version 1995, Berkeley, CA, USA).

Results

Subject data

At baseline, the mean number of remaining teeth in the 16 subjects was 26.0 ± 2.3, and a mean of $50.7 \pm 23.1\%$ of the teeth exhibited PD ≥ 5.0 mm. The mean percentage of sites with various clinical conditions was as follows: periodontitis sites for 20.3 ± 13.1, gingivitis sites for 44.9 ± 13.7 and clinically healthy sites for 34.8 ± 18.9. Healthy (1 site), gingivitis (1–2 sites) and periodontitis sites (3–4 sites) were selected for investigation in each subject, apart from two subjects in whom there was no appropriate and accessible healthy site to be sampled. A total of 85 sites were investigated both at baseline and 1 month after the periodontal treatment, consisting of 14 healthy sites, 17 gingivitis sites and 54 periodontitis sites.

On the basis of subject, mean percentage of sites with detectable plaque (PI ≥ 1) was significantly reduced from $90.3\% \pm 13.9\%$ at baseline to $58.5\% \pm 34.1\%$ after the treatment ($p < 0.01$). The mean percentage of sites with BOP decreased from $70.2\% \pm 20.1\%$ to $44.0\% \pm 25.4\%$ ($p < 0.01$) accordingly. Overall, the mean PD decreased from 4.8 ± 1.5 mm at baseline to 3.8 ± 1.7 mm after the treatment ($p < 0.001$). For the 54 periodontitis sites, the mean PD was significantly reduced from 5.7 ± 1.0 mm to 4.5 ± 1.7 mm following the treatment ($p < 0.001$). Of these sites, 55.5% showed a significant reduction of PD ≥ 1.0 mm, 24.1% with a minor reduction of PD < 1.0 mm and 20.4% with no change or an increase of PD.

Prevalence of the target periodontopathogens

At baseline, *A. actinomycetemcomitans* could not be detected in any subjects, whereas it was found in three sites from three subjects after the treatment, with a mean PD of 4.8 ± 0.7 mm and range 4.0–5.2 mm. *B. forsythus, P. gingivalis* and *T. denticola* were present in all subjects, and *P. intermedia* was found in 94% of the subjects. As compared to baseline, the prevalence of *B. forsythus* and *T. denticola* in the subjects was significantly reduced 1 month after the treatment ($p < 0.01$). In the 85 sites sampled, *B. forsythus, P. gingivalis, P. intermedia* and *T. denticola* were detected in over 50% of the sites. At 1 month post-treatment, the detection frequency of these four species decreased significantly, compared to that at baseline ($p < 0.01$) (Table 1).

The various infection patterns of the five target species in the 16 subjects before and after the treatment are shown in Table 2. No sites were found with single presence of *A. actinomycetemcomitans* and *B. forsythus*, while the numbers of subjects and sites with single presence of the other three species were low, both at baseline and after the treatment. The predominant infection pattern was *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola* at baseline and its detection frequency was significantly reduced after the treatment, based on both subject and site levels ($p < 0.01$). In contrast, the prevalence of the sites without the target species increased significantly after the treatment ($p < 0.01$). The frequency of co-infection of *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola* decreased significantly at periodontitis sites after the treatment ($p < 0.01$). Meanwhile, the detection frequency of none of the target species increased significantly ($p < 0.01$) at either subject or site levels (Table 3).

Changes in IL-8 and granulocyte elastase levels in GCF

Overall, IL-8 and MR-EA levels in GCF were significantly reduced after scaling and root planing ($p < 0.001$) with corresponding reductions of total counts of the target species. Concerning various categories of the clinical sites, MR-EA levels were significantly reduced in periodontitis sites ($p < 0.01$) and IL-8 levels decreased significantly in gingivitis sites ($p < 0.01$) (Table 4).

Interrelations of change in detection of subgingival periodontopathogens to IL-8 and MR-EA levels

Of the 54 periodontitis sites in the 16 subjects at baseline, 36 sites (66.7%) in 15 subjects were co-infected by *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola.* Of these 15 subjects, eight showed at least one site without persistent co-infection of *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola*

Table 1. Subjects and individual sites with *A. actinomycetemcomitans* (*A.a.*), *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) and *T. denticola* (*T.d.*) at baseline and 1 month after scaling and root planing in the 85 sampled sites of 16 subjects with chronic periodontitis

	Subjects[a]		Sites	
Organisms	Baseline n (%)	1 month n (%)	Baseline n (%)	1 month n (%)
A.a.	0 (0)	3 (18.8)	0 (0)	3 (3.5)
B.f.	16 (100)	9 (56.3)*	53 (62.4)	15 (17.6)*
P.g.	16 (100)	14 (87.5)	62 (72.9)	30 (35.3)*
P.i.	15 (93.8)	11 (68.8)	46 (54.1)	24 (28.2)*
T.d.	16 (100)	8 (50.0)*	58 (68.2)	17 (20.0)*

[a]At least one site with bacteria $\geq 10^3$ cells.
Significant difference from baseline, $*p < 0.01$.

after the treatment, while six subjects showed at least one site with persistent co-infection of these four species after the treatment. As shown in Table 5, the group of subjects without persistent co-infection of the target species exhibited a significant decrease of total IL-8 ($p<0.02$) and MR-EA levels ($p<0.02$), and concomitant reduction in PD, from 5.6 ± 0.2 mm at baseline to 3.8 ± 0.2 mm at 1 month ($p<0.01$) as well as reduction in percentage of sampled sites with detectable plaque ($p<0.02$) and BOP ($p<0.02$). No such change was found in the other group of subjects with persistent co-infection of the species after the treatment. At baseline, the group of subjects without persistent co-infection of the four species after treatment had higher IL-8 levels than the subjects with persistent co-infection after treatment ($p<0.01$). After the treatment, the former had higher IL-8 concentrations ($p<0.02$), lower GCF volume ($p<0.02$) and lower percentage of sampled sites with BOP ($p<0.02$) as well as PD ($p<0.01$) than the latter.

Of the 19 sites with none of the target species at baseline in 12 subjects, 14 sites (10 healthy sites and four gingivitis sites) in 10 subjects continued to show undetectable levels of these species after treatment, with a corresponding reduction in IL-8 levels ($p<0.02$), MR-EA levels ($p<0.02$) and PD ($p<0.02$). The other five sites (one healthy site, one gingivitis site and three periodontitis sites) in five subjects harboured one to three species after the treatment with a corresponding reduction in IL-8 levels ($p<0.02$) but not in MR-EA levels and PD.

Table 2. Single or combined infection patterns of *A. actinomycetemcomitans* (*A.a.*), *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) and *T. denticola* (*T.d.*) at baseline and 1 month after scaling and root planing in the 85 sampled sites of 16 subjects with chronic periodontitis

	Subjects[a]		Sites	
Infection patterns	Baseline n (%)	1 month n (%)	Baseline n (%)	1 month n (%)
P.g.	3 (18.8)	5 (31.3)	3 (3.5)	5 (5.9)
P.i.	1 (6.3)	1 (6.3)	1 (1.2)	1 (1.2)
T.d.	1 (6.3)	1 (6.3)	1 (1.2)	1 (1.2)
B.f. & P.g.	4 (25.0)	1 (6.3)	4 (4.7)	1 (1.2)
B.f. & T.d.	2 (12.5)	1 (6.3)	2 (2.4)	1 (1.2)
P.g. & P.i.	0 (0)	6 (37.5)*	0 (0)	8 (9.4)*
P.g. & T.d.	7 (43.8)	0 (0)**	7 (8.2)	0 (0)*
A.a., P.g. & P.i.	0 (0)	1 (6.3)	0 (0)	1 (1.2)
B.f., P.g. & T.d.	1 (6.3)	1 (6.3)	3 (3.5)	1 (1.2)
P.g., P.i. & T.d.	2 (12.5)	2 (12.5)	1 (1.2)	2 (2.4)
B.f. P.g., P.i. & T.d.	15 (93.8)	6 (37.5)**	44 (51.8)	10 (11.8)**
A.a., B.f., P.g., P.i. & T.d.	0 (0)	2 (12.5)	0 (0)	2 (2.4)
None [b]	12 (75.0)	15 (93.8)	19 (22.4)	52 (61.2)**
Total			85 (100)	85 (100)

[a]At least one site with the infection patterns concerned.
[b]None of *A.a., B.f., P.g., P.i.* and *T.d.* ($<10^3$ cells).
Significant difference from baseline, *$p<0.05$, **$p<0.01$.

Table 3. Presence (%) of the predominant co-infection by *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) and *T. denticola* (*T.d.*) under various conditions at baseline and 1 month after scaling and root planing

	Healthy		Gingivitis		Periodontitis	
Infection patterns	Baseline n (%)	1 month n (%)	Baseline n (%)	1 month n (%)	Baseline n (%)	1 month n (%)
Subjects[a]						
B.f. P.g., P.i. & T.d.	1 (7.1)	0	6 (37.5)	1 (6.3)	15 (93.8)	6 (37.5)*
None[b]	9 (64.3)	11 (78.6)	5 (31.3)	9 (56.3)	3 (18.8)	11 (68.8)*
Sites						
B.f. P.g., P.i. & T.d.	1 (7.1)	0	7 (41.2)	1 (5.9)	36 (66.7)	11 (20.4)*
None	11 (78.6)	13 (92.9)	5 (29.4)	13 (76.5)	3 (5.6)	26 (48.1)*

[a]At least one site with the infection patterns concerned.
[b]None of *A.a., B.f., P.g., P.i.* and *T.d.* ($<10^3$ cells).
Significant difference from baseline, *$p<0.01$.

Interrelations of change in IL-8 levels to MR-EA and detection of subgingival periodontopathogens

For the 36 periodontitis sites co-infected by *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola* at baseline in 15 subjects, the treatment resulted in either a reduction of IL-8 levels (Group A, eight subjects) or an increase of IL-8 levels (Group B, seven subjects). It is shown from Table 6 that in Group A, a significant reduction of IL-8 levels was accompanied by a concomitant reduction in MR-EA ($p<0.02$), PD ($p<0.01$),% of sites with co-infection of the target species ($p<0.01$) and total count of the target species ($p<0.02$). In contrast, no significant change of MR-EA levels and PD was noted in Group B, although significant reduction was

Table 4. IL-8 and MR-EA levels (mean ± SE) in GCF in the 16 subjects under various conditions at baseline and 1 month after scaling and root planing

	Healthy		Gingivitis		Periodontitis	
Parameters	Baseline	1 month	Baseline	1 month	Baseline	1 month
GCF volume (μL)	0.25 ± 0.05	0.26 ± 0.06	0.52 ± 0.07	0.34 ± 0.10	0.57 ± 0.04	0.54 ± 0.06
Total IL-8 (pg/site)	75.62 ± 25.52	47.58 ± 29.89	66.63 ± 22.58	12.10 ± 6.64*	70.30 ± 19.41	38.62 ± 10.30
IL-8 concentration (pg/μL)	339.16 ± 120.32	244.31 ± 150.79	213.41 ± 69.97	46.54 ± 27.84*	126.69 ± 235.36	80.97 ± 31.37
MR-EA (mAbs/min/site)	0.15 ± 0.04	0.11 ± 0.06	2.14 ± 0.84	0.11 ± 0.04	3.14 ± 0.84	0.62 ± 0.21*

Significant difference from baseline, *$p<0.01$.

found in percentage of sites with co-infection of the target species ($p < 0.01$) and total count of the target species ($p < 0.02$). At baseline, Group A showed significantly higher IL-8 levels than Group B ($p < 0.01$). After the treatment, Group B had higher GCF volume ($p < 0.02$) and MR-EA levels ($p < 0.02$) as well as deeper PD ($p < 0.02$) than Group A. In terms of the presence of various infection patterns after the treatment, the percentage of sites with none of the target species was significantly higher in Group A than that in Group B after treatment ($p < 0.02$) (Fig. 1).

Discussion

Current understanding of the pathogenesis of periodontitis disease suggests that it is of a multifactorial nature, being a result of the complex interactions between pathogenic subgingival microorganisms and the host response (Page et al. 1997). Increasing knowledge of this complexity provides new opportunities for diagnostic strategies to be investigated (Offenbacher et al. 1993). It has been strongly suggested that detection of specific subgingival bacteria together with identification of specific substances in GCF, as a measure of host response, may develop useful co-biomarkers for identification of disease progression and monitoring of treatment responses (Lamster et al. 1994, Wolff et al. 1994).

In line with these considerations, we

Table 5. Clinical and GCF data (mean ± SE) in the periodontitis sites from six subjects with and eight subjects without persistent co-infection by *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) and *T. denticola* (*T.d.*) 1 month after scaling and root planing

	Persistent co-infection[a]		Without persistent co-infection[b]	
Parameters	Baseline	1 month	Baseline	1 month
Clinical				
PD	6.0 ± 0.4	5.8 ± 0.5	5.6 ± 0.2	3.8 ± 0.2**††
CAL	13.8 ± 0.8	14.3 ± 1.1	12.7 ± 0.6	12.4 ± 0.6
% of sites with PI ≥ 1	91.7 ± 8.3	91.7 ± 8.3	95.8 ± 4.1	63.5 ± 13.0*
% of sites with BOP	91.7 ± 8.3	100.0 ± 0.0	84.4 ± 8.1	47.9 ± 13.5*†
GCF				
GCF volume (μL)	0.74 ± 0.08	0.85 ± 0.10	0.55 ± 0.07	0.37 ± 0.06†
Total IL-8 (pg/site)	21.67 ± 9.43	35.15 ± 13.74	89.22 ± 31.88#	47.62 ± 15.51*
IL-8 concentration (pg/μL)	31.34 ± 12.29	44.46 ± 17.09	153.42 ± 50.6#	129.49 ± 60.59†
MR-EA (mAbs/min/site)	1.92 ± 1.39	1.52 ± 0.92	4.12 ± 2.11	0.28 ± 0.20*

[a]Presence of co-infection by *B.f., P.g., P.i.* and *T.d.* at baseline and persistent co-infection of *B.f., P.g., P.i.* and *T.d.* 1 month after the treatment in at least one site of six subjects.
[b]Presence of co-infection by *B.f., P.g., P.i.* and *T.d.* at baseline, while absence of *B.f., P.g., P.i.* and *T.d.* 1 month after the treatment in at least one site of eight subjects.
Significant difference from baseline, *$p < 0.02$; **$p < 0.01$.
Significant difference from the subjects with persistent co-infection at baseline, #$p < 0.01$.
Significant difference from the subjects with persistent co-infection at 1 month, †$p < 0.02$; ††$p < 0.01$.

Table 6. Clinical, microbiological and GCF data (mean ± SE) at the periodontitis sites co-infected by *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) and *T. denticola* (*T.d.*) from Group A and Group B before and after scaling and root planing

	Group A[a]		Group B[b]	
Parameters	Baseline	1 month	Baseline	1 month
Clinical				
PD	6.1 ± 0.3	4.2 ± 0.6**	6.1 ± 0.5	5.7 ± 0.7†
CAL	12.5 ± 0.7	12.2 ± 0.7	12.8 ± 0.7	13.1 ± 0.9
% of sites with PI ≥ 1	81.3 ± 7.3	65.6 ± 9.2	100.0 ± 0.0	64.3 ± 17.9
% of sites with BOP	82.3 ± 9.5	41.7 ± 13.7	95.2 ± 4.8	61.9 ± 18.4
Microbiological				
% of sites with none of species	0	54.2 ± 16.6*	0	19.5 ± 10.6†
% of sites with 1–3 species	0	27.1 ± 12.6	0	50.5 ± 18.0*
% of sites with ≥ 4 species	100.0 ± 0.0	18.8 ± 13.2**	100.0 ± 0.0	30.0 ± 15.3**
Total count of 4 species	$8.9 \times 10^4 \pm 3.2 \times 10^4$	$3.3 \times 10^4 \pm 2.1 \times 10^4$*	$1.0 \times 10^5 \pm 2.6 \times 10^4$	$1.9 \times 10^4 \pm 1.4 \times 10^4$*
GCF				
GCF volume (μL)	0.65 ± 0.06	0.46 ± 0.09	0.50 ± 0.06	0.76 ± 0.11†
Total IL-8 (pg/site)	111.58 ± 35.81	31.83 ± 16.38**	14.14 ± 3.66#	58.13 ± 4.77*
IL-8 concentration (pg/μL)	174.57 ± 55.83	92.66 ± 64.97*	40.94 ± 16.67#	83.53 ± 11.01*
MR-EA (mAbs/min/site)	3.99 ± 1.74	0.10 ± 0.02*	1.10 ± 0.73	1.66 ± 0.74†

[a]Eight subjects with decreased levels of IL-8 after the treatment.
[b]Seven subjects with increased levels of IL-8 after the treatment.
Significant difference from baseline, *$p < 0.02$; **$p < 0.01$.
Significant difference from Group A at baseline, #$p < 0.01$.
Significant difference from Group A at 1 month, †$p < 0.02$.

recently examined the association between the presence of subgingival periodontopathogens and the resulting local inflammatory response (Jin et al. 1999, 2000a). Our data showed that there was a great variation in bacteria–host interactions among sites with different periodontal conditions in untreated chronic periodontitis. Although similar co-infections of *B. forsythus, P. gingivalis*, *P. intermedia* and *T. denticola* were frequently detected in the untreated periodontitis sites, there was a wide variation in host inflammatory response. Therefore, a more thorough evaluation of the risk for active periodontal disease may involve combined approaches to test the dynamic bacteria-host interrelations (Jin et al. 1999). The present study further evaluated the effect of scaling and root planing on the interrelations of subgingival periodontopathogens and IL-8 levels in GCF and crevicular levels of granulocyte elastase as a marker of intracrevicular granulocyte activity, and assessed their relations to the short-term treatment response among a group of chronic periodontitis patients who had not received any prior periodontal treatment.

Microbiological diagnosis, focussed upon a selected number of putative periodontopathogens, e.g. *A. actinomycetemcomitans*, *B. forsythus, P. gingivalis*, *P. intermedia* and *T. denticola*, has been proposed to be a useful tool for the identification of susceptible individuals and monitoring of disease progression (Dahlén 1993, Wolff et al. 1994). In the present study, alternations in microbiological profiles following the non-surgical periodontal treatment were expected, as mechanical debridement has been shown to induce beneficial changes in composition of the subgingival microflora (Lowenguth & Greenstein 1995), particularly *P. gingivalis, B. forsythus* and *T. denticola* (Haffajee et al. 1997a, b, Cugini et al. 2000). Overall, the present data showed that the detection frequency for the predominant co-infection with *B. forsythus, P. gingivalis*, *P. intermedia* and *T. denticola* was significantly reduced after the non-surgical periodontal treatment. The treatment had resulted in good reductions in probing depth and reductions in plaque and bleeding on probing, which, while substantial, were not yet the best that can be achieved. In contrast, the prevalence of sites without the target species being detected greatly increased. Those pockets with the persistence of infection by the target species showed no improvement in clinical parameters, as opposed to the sites with suppression of the co-infection of these species. It is conceivable that reduction in the prevalence of the subgingival periodontopathogens was related to favourable treatment responses, which agrees with previous studies (Haffajee et al. 1995, 1997b).

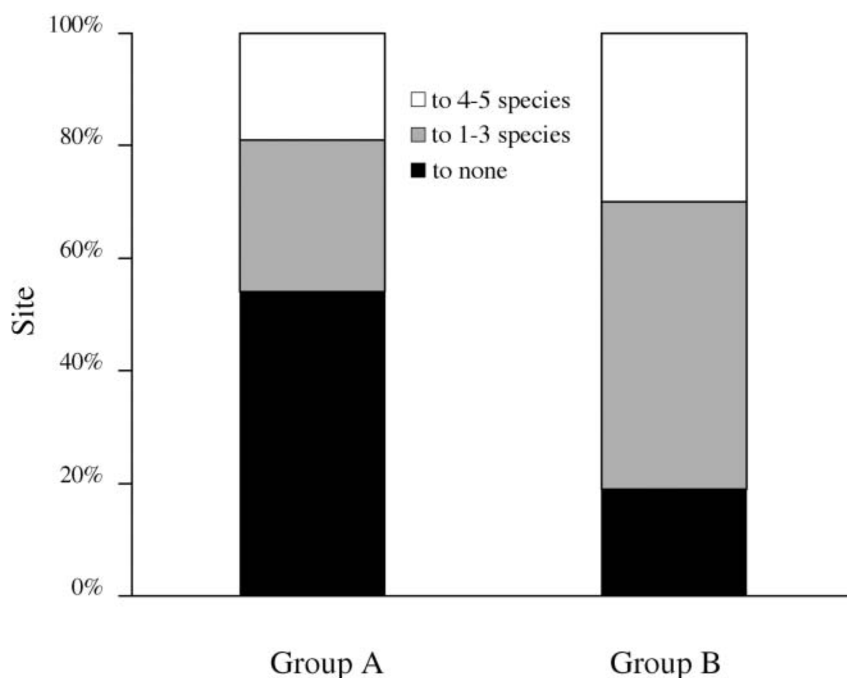

Fig. 1. The presence of various infection patterns after the treatment at the periodontitis sites co-infected by *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola* at baseline showing either decreased levels of IL-8 (Group A) or increased levels of IL-8 (Group B) after the treatment. The percentage of sites where none of the target species were detected after the treatment, i.e. the sites co-infected by *B. forsythus*, *P. gingivalis*, *P. intermedia* and *T. denticola* at baseline with none of these species detected after the treatment (four species to none), was significantly higher in Group A (54.2%) than that in Group B (19.5%) ($p<0.02$).

In previous studies, the effect of scaling and root planing on the target host-response markers in GCF has been examined. The present results of elastase levels in GCF are in accord with those reported by Cox & Eley (1992). Different results in terms of the effect of periodontal treatment on IL-8 levels in GCF have been reported (Tsai et al. 1995, Chung et al. 1997, Gamonal et al. 2000). In the present study, scaling and root planing generally reduced the levels of IL-8 and granulocyte elastase in GCF, with a corresponding reduction of total count of the target periodontal species. The possible explanations for the difference among these studies could be attributed to several factors, e.g. patient and site selection, sampling method employed, timing of clinical reassessment and possible variations in periodontal treatment protocol and treatment outcomes achieved.

Periodontal pathogenesis is characterized by complex interactions between pathogenic subgingival microorganisms and the host response (Page et al. 1997). The host immuno-inflammatory response to the challenge of periodontopathogens involves a complicated series of molecular events through a family of pattern recognition pathways in cell stimulation, e.g. LPS–LPS binding protein-CD14 and toll-like receptors (Aderem & Ulevitch 2000, Darveau 2000, Jin & Darveau 2001). Basic periodontal treatment is mainly designed to remove bacterial plaque, enhance plaque control and reduce periodontal inflammation. The present study examined the effect of basic periodontal therapy on the interrelationships between IL-8 related granulocyte elastase activity and the selected subgingival periodontopathogens in relation to the clinical treatment response. We found that after therapy those responding pockets without the detectable levels of the target species showed corresponding reductions in total IL-8

levels and elastase activity, while no such change was observed in the non-responding pockets with persistent co-infection of the target species. On the other hand, different patterns of change in elastase activity and probing depth were also observed in those sites with none of the target species at baseline but showing different infection patterns after the treatment, either remaining negative for the target species or harbouring the species. Taken together, these results indicate that persistence infection of the periodontopathogens is related to the locally unresolved inflammation, which may compromise the healing potentials and hence treatment outcomes. These observations are in accord with the general understanding of periodontal pathogenesis.

IL-8 has been shown to be important for the initiation and development of inflammation and tissue destruction in periodontal diseases, due to its critical role in recruitment and functional activation of neutrophilic granulocytes (Bickel 1993, Tsai et al. 1995, Chung et al. 1997, Gamonal et al. 2000, Jin et al. 2000a). Elevated IL-8 levels together with immense neutrophil accumulations have been reported in a number of human pathologic conditions characterized by inflammation and neutrophil infiltration, such as rheumatoid arthritis (Seitz et al. 1991) and acute respiratory distress syndrome (Matsumoto et al. 1997). However, little is known about the interrelationships of change in IL-8 levels following periodontal treatment to granulocyte elastase activity and infection patterns of subgingival periodontopathogens, and their potential implications in prediction and assessment of treatment outcomes.

Our recent studies show that the local host response to a similar co-infection of periodontopathogens in untreated periodontal pockets is diverse in terms of the intensity of inflammatory response measured by IL-8-associated granulocyte elastase activity in GCF. Moreover, there is a positive correlation between IL-8 levels and granulocyte elastase in GCF at periodontitis sites infected by the subgingival periodontopathogens (Jin et al. 2000a). The clinical implication of these findings should be further investigated. IL-8 and granulocyte elastase (as a marker of intracrevicular granulocyte activity) are closely related molecules. IL-8 plays an important role in the maintenance of local host-parasite equilibrium and in the limitation of neutrophil-associated tissue destruction (Tonetti et al. 1998). Under normal conditions, IL-8 expression relates well to the pattern of neutrophil infiltration, and appropriate release of IL-8 contributes to eliminating the infecting bacteria by neutrophils. Conversely, an uncontrolled release of IL-8 and resultant hyperactivity of neutrophils may cause tissue destruction (Yoshimura et al. 1997). In the present study, scaling and root planing yielded different outcomes relating to IL-8 and granulocyte elastase activities in GCF. Following treatment, IL-8 levels were reduced with a corresponding decrease in granulocyte elastase activity and reduction of probing depth, while those pocket sites with increased IL-8 levels after treatment exhibited different profiles of granulocyte elastase activity and healing responses. Accordingly, more favourable shifts in subgingival infection patterns were found in the former sites, as compared to the latter, although the total bacterial loading of the target species was significantly reduced in both groups of pocket sites after the treatment. IL-8 attracts neutrophils in a manner analogous to C5a and LTB_4, but IL-8-elicited activation pathways in the neutrophil are different. IL-8 can induce the release of matrix metalloproteinase-8 by neutrophils (Tonetti et al. 1993), a potent collagenase for degrading host connective tissues at sites of inflammation. It has been hypothesized that IL-8-elicited neutrophils in periodontitis or during acute abscess formation may have a different phenotype from that elicited by bacterial chemotactic peptides. The former phenotype would result in a tissue-destructive neutrophil phenotype, as opposed to the latter, which would be a more antimicrobial neutrophil (Offenbacher 1996). It seems that the dynamic interrelationship of IL-8-associated neutrophil activity to bacterial infection patterns is related to short-term treatment response. We speculate that the responding sites which showed probing depth reductions might show a positive neutrophil phenotype elicited by bacterial chemotactic peptides and balanced IL-8 and neutrophil interactions following the treatment as well as a favourable shift in bacterial infections, while the poor responding sites might reflect an IL-8-induced, tissue-destructive neutrophil phenotype through enhanced interactions of inflammatory mediators. At sites of inflammation, other inflammatory cytokines such as IL-1 can significantly enhance elastase release by IL-8-activated neutrophils (Brandolini et al. 1996, Owen et al. 1997) and, moreover, the neutrophil elastase released in the inflamed tissues converts IL-8 to more potent forms to enhance IL-8 activity (Padrines et al. 1994).

It was noted that the pocket sites that demonstrated an increase in IL-8 levels after treatment exhibited significantly lower baseline IL-8 levels in GCF than those pocket sites that showed a drop in IL-8 levels in GCF after treatment. In contrast to the former, the latter exhibited a corresponding reduction in granulocyte elastase activity and probing depth after the treatment, although all these pocket sites showed similar baseline clinical conditions and co-infection of the target species. These results suggest that for pocket sites the variations in initial IL-8 levels in GCF may implicate different short-term treatment outcomes, i.e. those pockets showing relatively high GCF IL-8 levels might indicate a more positive treatment response than those showing low GCF IL-8 levels before treatment.

Moreover, after the treatment, the remaining pocket sites also showed higher elastase activity than the responding pocket sites. Elevated elastase activity has been related to increased risk of attachment loss (Palcanis et al. 1992, Armitage et al. 1994, Renvert et al. 1998). Regulatory immuno-inflammatory response mechanisms to bacterial stimulus may result in complex interaction networks among cytokines and cellular components, producing inflammation and tissue destruction (Wilson et al. 1998). Based on the present results, it could be postulated that the continuous interactions between the co-infecting periodontopathogens and the IL-8-related granulocyte activity might result in a compromised healing process and maybe even increase the risk for further tissue destruction. The details of the mechanisms and their clinical implications warrant further investigation through long-term study.

The ability to determine treatment response and risk for future disease progression at pocket sites is an attractive and important issue with respect to assessing the merits of probing depth reduction. It has been shown that, after therapy, deep probing pocket sites are generally at greater risk of disease progression than shallow sites, while indi-

vidual probing depths are not good predictors of future attachment loss (Greenstein 1997). At present, no established parameter is available to differentiate the potentially destructive pockets from the relatively stable pockets. In this regard, it seems worthwhile to evaluate the relationship of change in host inflammatory response markers to the infection patterns of subgingival periodontopathogens as well as their links with treatment responses. Based on the present results, it seems that varying initial IL-8 levels in GCF and a corresponding shifting change of granulocyte elastase activity in GCF may characterize the different short-term treatment responses in untreated periodontal pockets. Hence, such dynamic interrelations might be indicative of various levels of risk for further disease progression at the remaining pocket sites. However, due to the limited sample size of the study and the short-term follow-up observation, further longitudinal studies are necessary to determine the clinical values of the findings.

Acknowledgements

We are grateful to Ms Carman Kong and Ms Mandy Chan for the clinical assistance. This study was supported by the Hong Kong Research Grants Council (RGC), Hong Kong SAR Government, the Committee for Research and Conference Grants (CRCG) of the University of Hong Kong, and the Karolinska Institute, Sweden.

Zusammenfassung

Zusammenhang zwischen Veränderungen der Interleukin-8-Spiegel und Ganulozyten-Elastaseaktivität in Sulkusflüssigkeit und subgingivalen Parodontalpathogenen nach nicht-chirurgischer Parodontitistherapie von Patienten mit chronischer Parodontitis

Zielsetzung: Untersuchung der Wirkung von Scaling und Wurzelglättung (SRP) auf die Wechselwirkungen zwischen subgingivalen Parodontalpathogenen und Interleukin-8 (IL-8) sowie Granulozyten-Elastaseaktivität in der Sulkusflüssigkeit (SF) und die Bestimmung ihrer Beziehung zu den kurzfristigen Resultaten der Therapie chronischer Parodontitis.

Material und Methoden: SF und subgingivale Plaque wurden bei 16 Personen mit unbehandelter chronischer Parodontitis vor und 4 Wochen nach SRP gesammelt. Die IL-8-Spiegel wurden mittels ELISA bestimmt. Die Granulozyten-Elastaseaktivität wurde mit einem spezifischen Substrat, pGluProVal-pNA, analysiert und die maximale Rate der Elastaseaktivität (MR-EA) berechnet. 5 DNS-Sonden wurden benutzt, um das Vorkommen von *A. actinomycetemcomitans* (*A.a.*), *B. forsythus* (*B.f.*), *P. gingivalis* (*P.g.*), *P. intermedia* (*P.i.*) und *T. denticola* (*T.d.*) mit einer Nachweisgrenze von 10*3* Zellen/Papierspitze nachzuweisen.

Ergebnisse: IL-8- und MR-EA-Spiegel in der SF nahmen nach SRP statistisch signifikant ($p<0{,}001$) ab, während sich die Zahl der Parodontalpathogene entsprechend reduzierte. Von den Stellen mit Sondierungstiefen (ST)= 5,0 mm und Koinfektion durch *B.f., P.g., P.i.* und *T.d* vor Therapie zeigten die Stellen, bei denen diese Koinfektion nach SRP nicht persistierte, eine signifikante Reduktion der IL-8- ($p<0{,}02$) sowie MR-EA-Spiegel ($p<0{,}02$) und der ST ($p<0{,}01$). Diese Veränderungen konnten an Stellen, an denen die Koinfektion persistierte, nicht festgestellt werden. Darüber hinaus war eine Reduktion der IL-8-Spiegel in den betreffenden Taschen von einer gleichzeitigen Reduktion der MR-EA-Spiegel ($p<0{,}02$) und der ST ($p<0{,}01$) begleitet, während keine signifikante Veränderung der MR-EA-Spiegel und der ST an Stellen beobachtet wurde, an denen die IL-8-Spiegel nach SRP anstiegen. Vor Therapie hatte die erste Gruppe von Stellen statistisch signifikant höhere IL-8-Spiegel aufgewiesen als letztere Gruppe ($p<0{,}02$).

Schlussfolgerung: Die IL-8-bezogene Granulozyten-Elastaseaktivität stand in Zusammenhang mit der Veränderung des Infektionsmusters der untersuchten Parodontalpathogene nach SRP. Unterschiedliche IL-8-Ausgangsspiegel in der SF und eine entsprechende Veränderung der Granulozyten-Elastaseaktivität in der SF könnten die verschiedenen kurzfristigen Effekte nicht-chirurgischer Therapie bestimmen.

Résumé

Relation entre les variations du taux d'interleukine-8 et de l'activité de l'élastase granulocytaire dans le fluide gingival créviculaire, et les parodontopathogènes sous-gingivaux après traitement parodontal non chirurgical chez des sujets atteints de parodontite chronique

Buts: Cette étude avait pour but de déterminer les effets du détartrage et du surfaçage radiculaire (*scaling and root planing*, SRP) sur les interactions entre les parodontopathogènes sous-gingivaux d'une part, et l'interleukine-8 (IL-8) ainsi que l'activité de l'élastase granulocytaire dans le fluide gingival créviculaire (*gingival crevicular fluid*, GCF) d'autre part. Son objectif consistait également à évaluer leur relation avec la réponse au traitement à court terme dans la maîtrise de la parodontite chronique.

Matériaux et méthodes: Le GCF et la plaque sous-gingivale ont été prélevés lors de l'examen initial puis quatre semaines après SRP sur 16 sujets atteints de parodontite chronique non traitée. Les taux de IL-8 ont été déterminés par la méthode ELISA. L'activité de l'élastase granulocytaire a été analysée avec un substrat spécifique, pGluProVal-pNA, et le taux maximal d'activité de l'élastase (*maximal rate of elastase activity*, MR-EA) a été calculé. Cinq échantillons d'ADN ont été utilisés pour détecter la présence de *A. actinomycetemcomitans (A.a.), B. forsythus (B.f.), P. gingivalis (P.g.), P. intermedia (P.i.)* et *T. denticola (T.d.)*, avec une sensibilité de 103 cellules/point de papier.

Résultats: Les taux de IL-8 et MR-EA dans le GCF ont diminué de façon significative après SRP ($p<0{,}001$), parallèlement à une réduction de la numération totale des espèces. Parmi les sites présentant une profondeur au sondage (*probing depth*, PD) de 5,0 mm et une co-infection par *B.f., P.g., P.i. et T.d.* au départ, ceux sans co-infection persistante par ces espèces après SRP affichaient une réduction significative des taux de IL-8 ($p<0{,}02$) et MR-EA ($p<0{,}02$), et de PD ($p<0{,}01$). Aucune variation de ce type n'a été observée au niveau des sites où une telle co-infection persistait. En outre, la baisse des taux de IL-8 sur ces sites s'accompagnait d'une réduction concomitante de MR-EA ($p<0{,}02$) et PD ($p<0{,}01$), alors qu'aucune variation significative des taux de MR-EA ni de PD n'était observée dans les poches présentant une hausse des taux de IL-8 après SRP. Lors de l'examen initial, les taux de IL-8 étaient significativement plus élevés dans le premier groupe de sites que dans ce dernier ($p<0{,}02$).

Conclusions: L'activité de l'élastase granulocytaire liée à l'IL-8 était en rapport avec la modification des types d'infection des parodontopathogènes cibles après détartrage et surfaçage radiculaire. Les fluctuations des taux initiaux de IL-8 dans le GCF et les variations correspondantes de l'activité de l'élastase granulocytaire dans le GCF pourraient caractériser les différentes réponses au traitement à court terme.

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