Case Report

Characterization of Patients Presenting for Treatment to a University Refractory Periodontal Diseases Unit: Three Case Reports

Vinay M. Bhide,* Howard C. Tenenbaum,*,† and Michael B. Goldberg*

Background: Of the many forms of periodontal disease, refractory periodontal diseases are the least characterized. They are defined as the continued degeneration of the periodontium despite adequate management. This has led to the suggestion that there may be a systemic component that is a contributing factor to the development of this condition. The objectives of this report were to follow the progression of clinical changes associated with periodontal disease over a number of years in this unique population and review various hematologic and microbiologic factors that may be contributing to the disease progression.

Methods: Three subjects were profiled. They were referred to the Refractory Periodontal Disease Unit at the University of Toronto by periodontists or general practitioners in the Southern Ontario region. Complete medical and dental histories were obtained along with baseline clinical measurements. Periodontal examinations were facilitated with the use of a computer-assisted periodontal probe. A microbiologic analysis using immunofluorescence techniques was able to detect Prevotella intermedia, Porphyromonas gingivalis, Tannerella forsythia, and Actinobacillus actinomycetemcomitans and spirochetes. A hematologic analysis, including a complete blood count (CBC), immune profile, and glycosylated hemoglobin assay, was also performed.

Results: The clinical presentation revealed that patients receiving adequate maintenance therapy and with good to excellent oral hygiene demonstrated sites with continual loss of attachment. Few periodontal pathogens were detected. However, the most significant finding appeared to be the report elevated levels of CD8+ cells within this group of patients compared to normal laboratory ranges.

Conclusions: This report is an attempt at characterizing a unique population within the periodontal realm. The long-term monitoring of these patients allowed for an assessment of factors that may be involved in the continued decline of the periodontal health of these patients. Based on the immune profile, it is possible that a hyperresponsive state may be the primary feature of this population. Future assessments, including full-mouth interleukin (IL)-1 and matrix metalloproteinase (MMP)-8 levels, may assist in characterizing this population further, with the goal of producing markers that will assist clinicians in predicting treatment outcome. J Periodontol 2006;77:316-322.

KEY WORDS
Immunology; microbiology; periodontal diseases, refractory; treatment.

Periodontal diseases comprise a variety of complex bacterial infections, which frequently occur as inflammatory and destructive lesions of the periodontium. Periodontitis is prevalent in humans and is characterized by degradation of soft and mineralized connective tissues. If allowed to progress, periodontitis has been shown to be the most common cause of tooth loss in adults.1 The clinical manifestations of most forms of periodontitis are sufficient to permit an accurate diagnosis when evaluated in conjunction with a thorough history and clinical examination.2 However, diagnosing periodontitis can be complicated due to the alternating active and quiescent phases of the disease and the requirement for repeated treatments over a long period of time. Furthermore, a tremendous variation of severity can exist within the same subject. These features of periodontitis are indicative of the need to develop diagnostic modalities, which extend beyond the traditional classification of disease based solely on oral clinical signs and symptoms.3

Of the different categories of periodontal diseases, refractory periodontal diseases have received some attention recently in the literature.4 Although different forms of chronic and aggressive periodontal diseases have been studied and characterized extensively, refractory periodontitis as a periodontal disease entity is

* Faculty of Dentistry, University of Toronto, Toronto, ON.
† Department of Laboratory Medicine and Pathobiology, Faculty of Medicine, University of Toronto.

doi: 10.1902/jop.2006.050108
still not clearly understood. Refractory periodontal diseases are characterized by continued degeneration of the periodontium despite ongoing sanative, surgical, and/or pharmacological therapy. This has led investigators to suspect that perhaps a systemic component is more influential to the development of refractory periodontal diseases than otherwise thought. A few studies have been reported in the literature attempting to elucidate the association between host immune response and refractory periodontal diseases. In particular, the role of T lymphocytes as a direct mediator in the immune response as well as its potential role in the production of inflammatory cytokines has been considered. Other factors such as the role of serum lipids, hyperresponsive monocytes/polymorphonuclear leukocytes (PMNs), and hyperresponsive macrophages have also been considered potential contributors to periodontal destruction.

However, improved screening methods and early diagnosis of patients more likely to develop refractory periodontitis will facilitate risk assessment and optimize treatment of persons at risk for this condition. These can only be made possible with a thorough understanding of clinical, microbiological, systemic, and immune parameters, their relationship, and how they are represented in current and potential periodontitis patients.

The objectives of this report were as follows: 1) to obtain long-term measurements of clinical parameters in patients presenting to a university-based refractory periodontal disease unit including probing depth (PD), bleeding on probing (BOP), tooth mobility, and percentage of sites with plaque and calculus; 2) to obtain an oral microbiologic profile of these patients with respect to common putative periodontal pathogens such as *Prevotella intermedia*, *Porphyromonas gingivalis*, *Tannerella forsythia*, and *Actinobacillus actinomycetemcomitans* and spirochetes; 3) to characterize these patients on the basis of the following markers of host immune response status: white blood cell count and associated components, CD4+, CD8+, and CD4:CD8 ratio; and 4) to assess the patient’s status with respect to blood sugar levels and chronic levels by measuring the amount of glycated hemoglobin (HbA1c); and 5) to assess the patient’s potential genetic susceptibility to periodontal disease based on the genotype characteristic related to interleukin-1 alpha (IL-1α) and beta (IL-1β) using the periodontal susceptibility test (PST).

**METHODS**

Patients who were deemed to have refractory periodontitis were referred to the Severe and Refractory Periodontal Disease Research and Treatment Unit, a subgroup of the University of Toronto’s Dental Research Institute. All patients were referred to this unit by general practitioners or specialists in periodontics in the Southern Ontario region and had been treated previously for periodontitis using either surgical or non-surgical modalities.

A complete medical history, dental treatment history, and baseline measurements including full-mouth PD were obtained using a periodontal probe for each subject. Included in the examination were the presence of BOP for buccal and lingual surfaces of each tooth and the number of mobile teeth and sites with plaque/calculus.

A detailed microbiologic profile was obtained for each patient. The oral microbiologic analysis was done to determine levels of commonly known putative periodontal pathogens such as *P. intermedia* (*Pi*), *P. gingivalis* (*Pg*), *T. forsythia* (*Tf*), and *A. actinomycetemcomitans* (*Aa*) and spirochetes. Crevicular fluid samples from the mesio-buccal pockets of teeth #3, #9, #19, and #25 were obtained for analysis using paper points.

The levels of the various periodontal pathogens were measured using immunofluorescence techniques. Hematologic analysis was performed in conjunction with the Department of Laboratory Medicine and Pathology, Mount Sinai Hospital, Toronto. This analysis included total white cell count and its associated individual components (i.e., neutrophil, lymphocyte, monocyte, eosinophil, and basophil) presented in absolute numbers and fractional percentages. Additionally, the numbers and percentages of CD3 (data not shown), CD4, and CD8 and the CD4:CD8 ratio were analyzed.

Once the information from the initial assessment was gathered, treatment recommendations were made to the referring dentist. Recommended treatment included scaling and root planing with local anesthetic, systemic antimicrobial therapy, subantimicrobial dose doxycycline (SDD), and bisphosphonate therapy. Patients were seen for follow-up in our clinic with a repeat of the parameters taken. If patients demonstrated continued attachment loss with this mode of care, they were deemed to be refractory to treatment.

The three patients described here were examined in our clinic, semiannually, from 1998 until the present. The clinical and hematologic parameters trace the long-term results of these patients’ maintenance history. Prior to presenting to our clinic, each patient underwent sanative therapy and antimicrobial treatment. Surgical therapy was also performed in two of the cases. Informed consent was obtained for all treatment and evaluation procedures rendered for the patients described in this report. The investigation was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2000.
CASE 1
AW is a 33-year-old female who presented for assessment in the Severe and Refractory Periodontal Disease Research Unit. She had been seeing a periodontist since 1993. At that time, she underwent periodontal scaling and root planing with local anesthetic and sedation. Periodontal flap surgery in the four posterior sextants and three mucogingival procedures in the anterior region were performed in 1995. Since those treatments, she had numerous courses of antibiotic therapy, including amoxicillin and metronidazole. She was a non-smoker. The concern of the referring periodontist was the continued attachment loss, noted both clinically and radiographically (Fig. 1) despite superlative home care and regular maintenance every 3 months.

Upon presentation, the clinical examination indicated 34% of probing depths >4 mm. However, only 8% of sites demonstrated bleeding on probing. It was also noted that 26 of 28 teeth demonstrated recession ≥2 mm, with a range of 2 to 4 mm. In addition, supragingival plaque was found in only seven of 168 sites. The microbiologic assessment showed that the mesio-buccal side of tooth #3 was the only site harboring any periodontal pathogen (P. intermedia), and even then, the levels were classified as being very low (Fig. 4A). The percentage of CD8+ levels was elevated above the normal range (Fig. 2). CD4+ levels were within a normal range. AW was found to be PST positive. The mean glycated hemoglobin level, measured from 1999 until the present, was 5.2% ± 0.07%.

AW was placed on SDD and bisphosphonates along with continued 3-month recalls of scaling and root planing under sedation. Despite this treatment, probing depth and attachment loss continued to progress (Fig. 3). Bacterial deposits were minimal, as noted in Figure 4. However, CD8+ levels remained elevated relative to maximal baseline values.

CASE 2
WB is a 44-year-old female who has a past history of severe bulimia and malnutrition. However, WB maintained a constant weight and has been symptom free since 1994. She never smoked. WB saw a periodontist every 3 months since 1996. The periodontist’s primary concern was that of continued recession despite adequate maintenance for many years. She had no past history of periodontal surgical intervention.

Although probing depth remained relatively shallow and constant, WB noted significant and progressive recession since 1999, as noted in Figure 5. Microbiologic deposits were minimal. However, the CD8+ level was elevated relative to the normal maximal range (Fig. 6). In addition, yeast hyphae on the tongue and gingiva were noted. WB was found to be PST negative. The mean glycated hemoglobin level was 5.0% ± 0.02% as measured over the course of treatment.

Past treatment for WB included mycostatin 100,000 IU rinse to address the candida superinfections. Also,
she maintained a 3- to 4-month recall schedule with her periodontist. Despite the low levels of pathogens and the excellent oral hygiene, WB continued to lose attachment. She was placed on a dose of alendronate, 70 mg orally per week. Short courses of metronidazole, 500 mg three times a day, and amoxicillin, 500 mg three times a day, were administered when bacterial levels increased slightly. When the progression of attachment loss was noted, WB was placed on a regimen of SDD, which continues at the present time.

CASE 3
HP is a 43-year-old male who presented to the clinic with a diagnosis of severe, generalized chronic periodontitis. HP never smoked. HP had periodontal surgery performed in all six sextants, yet continued to demonstrate progressive bone loss (Fig. 7). Upon presentation to our clinic, immunofluorescence was unable to detect the presence of bacterial deposits. Initially, HP was placed on a course of SDD. CD8+ levels were elevated at each assessment and remained so throughout the study period. At one follow-up appointment, mild increases in P. gingivalis and P. intermedia were noted. HP was placed on a course of metronidazole, 500 mg three times a day,
plus amoxicillin, 500 mg three times a day for 7 days. Although bacterial counts returned to undetectable levels, progressive attachment loss continued. HP was PST negative. The mean glycated hemoglobin level was 5.2% ± 0.04% over the course of the study period.

In January 2004, a dental implant was placed due to tooth loss in the posterior mandible (Fig. 8). However, within 3 months of placement, crestal bone loss around the neck of the fixture occurred, bringing into doubt the prognosis of the implant.

**DISCUSSION**

The long-term outcome of periodontal therapy in a private practice setting has been reviewed, and it was concluded that there was a cohort of patients who, despite seemingly adequate therapy and faithful long-term dental maintenance, continued to show periodontal breakdown. Refractory periodontal diseases were once considered a separate and possibly more prevalent disease entity based on previous diagnostic categories. In fact, more recent definitions consider this condition as a subset of all periodontal disease categories not responsive to conventional therapies. The goal of this report was to identify characteristics unique to this population, insofar as these parameters may assist clinicians in long-term treatment planning and prognostication once they are identified.

Prior to identifying a case of periodontal disease as refractory to treatment, it is important to assure that all modalities of conventional therapy have been undertaken. Two of the three subjects reviewed here had at least one surgical therapy performed, and, in one case, all six sextants underwent surgery. Each subject was on a regular recall schedule and received supragingival and subgingival maintenance therapy. Additionally, a periodontist saw each patient every 3 months for recall maintenance. This form of treatment is consistent with that demonstrated by Westfelt et al. in that supra- and sub-gingival scaling was required to prevent further periodontal destruction in cases with advanced disease.

A review of the medical history was done for all subjects to confirm that a systemic disease was not
contributing to the progression of the condition. The glycated hemoglobin measurements,\textsuperscript{14} which were normal in all three cases, and immunodeficiency profiles\textsuperscript{15} that were performed were meant to rule out diabetes and HIV, respectively, as sources of systemic disease.

One seemingly unique variable is that of smoking. It has been known for some time that smoking is a major risk factor in the development of periodontal disease. It has been suggested that current smokers are four times more likely to develop periodontal disease compared to persons who never smoked.\textsuperscript{16} Yet, in our particular cohort of patients, none of the patients ever smoked, thereby eliminating this factor as an entity associated with the lack of response to therapy.

The involvement of T lymphocytes in periodontal bone destruction and the inflammatory lesion has been studied for some time. Evidence suggesting a host antibody response to the presence of $A_a$ is indicative of the fact that a T-cell response occurs in the presence of oral microorganisms.\textsuperscript{17} In particular, this study focused on T-cell receptor subtypes CD4\textsuperscript{+} and CD8\textsuperscript{+}. CD4\textsuperscript{+} cells are thought to be associated with the initiation of clonal expansion of B and T cells, resulting in a perpetuation of the inflammatory process. Lymphoid cells with the CD8\textsuperscript{+} cell receptor are thought to be natural killer cells. Although it has yet to be fully elucidated, there are many ways that these subgroups of cells may contribute to the activity of a periodontal lesion, including elevated production of IL-1.\textsuperscript{18}

Although it must be noted that the fractionated white blood count values for the study population were all within normal laboratory ranges, it is interesting to observe that the CD8\textsuperscript{+} values were elevated relative to the normal laboratory ranges. Although two of the three cases were PST negative, this still may suggest an inherent hyperresponsiveness of a refractory patient to the presence of plaque, such that excessive breakdown, possibly facilitated by the inflammatory mediators associated with this T-cell subset, may be occurring.

Although our report focused on T-cell subsets as potential markers for refractory periodontal diseases, other studies have demonstrated a possible role for serum lipids, PMN/monocyte, and macrophage function in association with tissue destruction. Recent evidence has suggested a bidirectional relationship between periodontal disease and hyperlipidemia, such that elevated serum lipid levels induce an increase in PMN production of proinflammatory cytokines such as IL-1B.\textsuperscript{19} Indeed, data currently under analysis in our treatment/research program suggest that patients with ongoing breakdown may actually have hypersensitive and hyperreactive PMN cells (data not shown). Also, a survey of all 50 patients being treated in the unit (albeit recognizing that some small proportion might not be refractory according to our criteria) confirmed that higher mean CD8\textsuperscript{+} levels were seen compared to the external control lab values. As for the elevated PMN activity observed in refractory patients, these cells were compared to PMNs from patients with periodontitis who responded to treatment and accordingly had PMNs with a more normal activity level and number.

From the data gathered, it may be possible to construct a profile of a patient who may fit the category of severe/refractory. The purpose of this would be to identify characteristics that may be unique to this population that may assist in early identification of a patient who may potentially experience severe or refractory disease that is unresponsive to conventional therapies. In doing so, a change in treatment strategy (i.e., extraction and implant therapy rather than further conventional periodontal therapy) may be considered early on in the case. However, in the case of subject 3 (HP), it is possible that even implant therapy may be more susceptible to breakdown in this unique population. Further work is being performed in the University of Toronto’s Dental Research Institute to determine if there is indeed an immune profile or characteristic that may make an individual more susceptible to periodontal breakdown and/or implant failure.

CONCLUSIONS

Given the limitations of this report, it is difficult to draw any firm conclusions because control patients (i.e., healthy and those with non-refractory disease) were not included. However, there appears to be a cohort of patients who demonstrate a continual decline in their periodontal condition despite adequate treatment and maintenance. Speculation varies widely as to the reason for this decline\textsuperscript{6,20} but tends to focus on the microbiologic aspect of disease. A new
concept introduced in 2002 suggested that some forms of refractory disease might present with progressive loss of attachment and alveolar bone, the absence of gingival inflammation or microbial deposits, and the failure of the disease to respond to traditional periodontal therapies. This condition was termed “non-inflammatory destructive periodontal disease.”

The mechanism by which refractory periodontal diseases progress requires further investigation. However, it is possible that patients who do not respond to conventional therapy may, in fact, be hyperresponsive hosts who react even to a small pathogenic bacterial load. It is possible that those with elevated CD8+ levels may show susceptibility to refractory periodontal diseases. Future studies will expand on identifying the levels of inflammatory mediators present in this population, including IL-1 and MMP-8 (samples already collected and being analyzed), such that further characterization of this population may provide for early recognition of future breakdown and modification of treatment approaches. Moreover, in the absence of adequate understanding of and treatments for refractory periodontal diseases, parameters that are currently under study in the Refractory Periodontal Disease Unit at the University of Toronto could lead to the identification of prognostic markers that would permit the clinician to direct a patient toward ongoing periodontal maintenance and intervention or perhaps to earlier implementation of endosseous implants. Furthermore, ongoing studies in this unit will focus on quality of life indices to determine the psychosocial effects of diagnosis, treatment, and daily management of refractory periodontitis. This could provide a database to compare the quality of life among other parameters in patients receiving ongoing periodontal intervention compared to similar patients treated sooner with endosseous implants.

REFERENCES


Correspondence: Dr. Michael B. Goldberg, Faculty of Dentistry, 124 Edward St., Toronto, ON M5G 1X5. Fax: 416/586-5010; e-mail: m.goldberg@utoronto.ca.

Accepted for publication June 16, 2005.