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Modulation of clinical expression of plaque-induced gingivitis

I. Background review and rationale

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Abstract

Objectives: The purpose of this article is to provide the necessary background and rationale for the accompanying studies, which are ultimately aimed at identifying genetic and environmental factors determining gingivitis susceptibility.

Materials and methods: The literature on factors reported to modify the clinical expression of gingivitis, i.e., factors that determine individual variability in gingival inflammatory response to plaque, is presented.

Results: Clinical evidence suggests that the gingival inflammatory response to plaque accumulation may differ substantially among individuals. However, most of the available studies are of small scale and not purposely designed to address the issue. Systemic factors implicated in modulation of the clinical expression of gingivitis include metabolic, genetic, environmental and other factors. The significance of such factors in designing and conducting a large-scale experimental gingivitis trial and means to account for them are discussed.

Conclusion: Although several factors have been implicated, genetic or environmental factors underlying differences in gingivitis expression are not fully elucidated. The accompanying studies aim to identify and characterize, among participants in a specifically designed large-scale experimental gingivitis trial, subjects that differ significantly in their gingival inflammatory response to plaque. This is the first step in an effort to determine genetic or environmental factors underlying such differences.

Key words: experimental design; gingivitis; gingivitis/etiology; gingivitis/physiopathology

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The two most prevalent and most investigated diseases of the periodontium, i.e. plaque-induced gingivitis, a reversible condition, and chronic periodontitis, an irreversible condition that can lead to tooth loss, are chronic inflammatory conditions an of an infectious nature (Williams 1990). The unequivocal role of dental bacterial plaque in the development of these diseases has been established since almost 40 years ago (Löe et al. 1965, Theilade et al. 1966, Lindhe et al. 1973, Slots & Hausmann 1979, Holt et al. 1988, Breuer & Cosgrove 1989, Persson et al. 1994). Although ample evidence suggests that susceptibility to periodontitis varies considerably among individuals (Löe et al. 1986), in large part because of genetic factors (Hart & Kornman 1997, Michalowicz et al. 2000), there are limited studies addressing a potential host-dependent variation in susceptibility to gingivitis.

Dental plaque-induced gingivitis (Mariotti 1999) is the most prevalent disease (Oliver et al. 1998, Albandar & Tinoco 2002, Sheiham & Netuveli 2002) among the numerous conditions that affect the periodontium (Armitage 1999, Laskaris & Scully 2003). The definitive evidence provided by the pioneering studies of Löe and colleagues in the mid 1960s confirmed that gingivitis is caused by dental bacterial plaque (Löe et al. 1965, Theilade et al. 1966). The etiologic role of plaque in the development of gingivitis is further supported by numerous intervention studies. Plaque reduction or elimination, either by mechanical means (Löe et al. 1965, Bosman & Powell 1977, Brecx et al. 1988) or chemical anti-microbial agents (Löe et al. 1967, Bosman & Powell 1977, Brecx et al. 1990), leads to disease resolution. Earlier, experimental gingivitis studies have alluded to possible subject differences in susceptibility to gingivitis. Such differences have been usually ascribed to differences in plaque accumulation rates (quantitative plaque differences) and/or differences in plaque species present (qualitative plaque differences) (Löe et al. 1965, Theilade et al. 1966). However, quantitative and/or qualitative plaque differences cannot account for all the reported differences in gingivitis susceptibility (Abbas et al. 1986a, Lie et al. 1995, 1998). The scientific evidence considered above is based on clinical studies.

The clinical assessment of the gingival inflammatory response to plaque accumulation relies on several parameters (Mariotti 1999). Visually assessed parameters include the extent and severity of changes in the physical status of the gingiva, such as changes in color, in surface anatomy (contour), and in bleeding tendency. Several indices have been proposed for the clinical evaluation of gingival inflammation, including the papilla, marginal, attached (PMA) index (Schour & Massler 1947), the papillary bleeding index (PBI) (Mühlemann 1977), and the gingival index (GI) of Löe & Silness (1963), among others (Ciancio 1986). The visually assessed signs of inflammation correspond to histopathological tissue changes (Hancock et al. 1979, Greenstein et al. 1981, Engelberger et al. 1983). The various indices have been shown to each have its own limitations, a major common one being the subjective nature of some of the required determinations. To add objective means to the clinical assessment of the status of the gingiva, quantitation of the gingival crevicular fluid has been developed and employed widely (Golub & Kleinberg 1976, Hinrichs et al. 1984a, b, Ciantar & Caruana 1998). Abundant evidence indicates that crevicular fluid flow correlates with both the clinical and the histological status of the gingiva (Oliver et al. 1969, Rüdin et al. 1970, Daneshmand & Wade 1976, Engelberger et al. 1983). Methods and indices to quantify the accumulated dental bacterial plaque, such as the Debris index (Greene & Vermillion 1960), the Quigley and Hein plaque index (Quigley & Hein 1962), and the plaque index by Silness & Löe (1964), among others (Fischman 1986), have been necessary in the assessment of the gingival response to the plaque. Use of these indices has helped demonstrate the relationship between the extent of plaque deposits and the severity of gingivitis (Breuer & Cosgrove 1989, Müller et al. 2000). Beyond the quantitative aspects of this relationship, there are important qualitative aspects, as well. Analysis of the microbial composition of plaque has led to the implication of certain bacterial species in the development of gingivitis (Moore et al. 1984). Furthermore, more sophisticated means to evaluate the inter-relationship between accumulated plaque and gingival response have been pursued from the qualitative aspect of the host response, such as characterization of relevant molecular determinants present in crevicular fluid (Lamster et al. 1985, Patters et al. 1989, Heasman et al. 1993a, Herrmann et al. 2001, Lundy et al. 2001).

Regardless of the methods used for the clinical evaluation of the gingiva (or plaque) in either natural or experimental gingivitis, it is clear that there are significant differences among individuals with respect to the severity and extent of gingivitis they experience. Characterization of the nature of these differences and identification of the underlying etiology have been the impetus for the design and implementation by the authors of a series of studies (Trombelli et al. 2003a, b, unpublished data) whose immediate goal is to define and characterize subpopulations with significant differences in gingivitis susceptibility. The ultimate goal of these studies is the identification of host-dependent variables, possibly genetic ones, which characterize the two subpopulations. The purpose of this review paper is to provide the necessary background and the rationale for these studies.

Factors modulating gingivitis expression

To undertake a series of large-scale experimental gingivitis studies aimed at identifying individual characteristics associated with increased susceptibility to plaque-induced gingival inflammation requires, at the very least, acknowledgement of the factors already established or implicated in the literature as modifiers of gingival inflammation. It also requires, whenever possible, measures to control for such factors, either during study design or during data analysis. In the following paragraphs, these factors are reviewed and discussed in relation to the design and execution of such an experimental gingivitis trial.

Local factors modulating gingivitis expression

In many circumstances, particularly in relation to natural gingivitis, the reasons for exacerbated manifestation of gingivitis have been attributed to local factors, implicated in increased plaque accumulation or hindrance of oral hygiene procedures. Such local factors reported to lead to deteriorating gingival/periodontal health include developmental or anatomical tooth variations [palato-gingival groove (Hou & Tsai 1993), enamel pearls (Goldstein 1979), crowding (Chung et al. 2000)], pathological tooth conditions [fractures (Polson 1977), caries (Albandar et al. 1995)], gingival anatomical conditions [recession defects (Smukler & Machtei 1987, Goutoudi et al. 1997), frenum position (Addy et al. 1987)], and iatrogenic factors [subgingival restoration margins (Waerhaug 1975, Bader et al. 1991), overhangs (Rodriguez-Ferrer et al. 1980, Lang et al. 1983), partial dentures (Bissada et al. 1974, Yeung et al. 2000), orthodontic appliances (Boyd & Baumrind 1992), etc.]. Maxillofacial anatomic variants (e.g. inadequate upper lip coverage) and/or upper respiratory obstructions (e.g. epipharyngeal adenoids, deviated nasal septum) may lead to mouthbreathing. Mouthbreathing and inadequate upper lip coverage have been shown to lead to changes in plaque accumulation and gingivitis expression, particularly in the maxillary anterior segment of the dentition (Jacobson 1973, Addy et al. 1987, Wagaiyu & Ashley 1991, Gulati et al. 1998).

The frequent intake of sucrose, acting on a local level, is well established as responsible for increased plaque accumulation and concomitantly increased gingival inflammation (Jalil et al. 1983, Sidi & Ashley 1984). Although addressed here, under local factors, because of its action, increased sucrose intake can also be considered an environmental factor.

Under the above conditions, the cause of the increased (or decreased) gingival inflammation manifested in the subjects is identifiable and amenable to corrective action. In the context of experimental gingivitis trial design, such factors are controllable by establishing appropriate inclusion and exclusion criteria, as well as detailed instructions for study participants. More importantly, such local factors are not expected to alter the inherent manner in which the host tissues respond to plaque accumulation, which is the main thrust of the present studies. Therefore, these local factors, although critical for individual patient management in daily practice, do not merit further discussion here.

Systemic factors modulating gingivitis expression

The most recent classification of periodontal diseases (Armitage 1999)

acknowledges that the clinical expression of plaque-induced gingival inflammation can be substantially modified by systemic factors, either inherent to the host or related to environmental influences (Mariotti 1999). For the purposes of this review, the systemic factors will be divided in the following categories: metabolic, genetic, environmental, and others. These systemic factors are summarized in Table 1.

Metabolic factors

Physiologic and pathologic endocrine changes have long been established as significant modifying factors in the expression of gingivitis (Sooriyamoorthy & Gower 1989). Among physiologic changes, the variation in sex hormone levels occurring during puberty (Mombelli et al. 1989, Bimstein & Matsson 1999) and pregnancy (Hugoson 1971) have been shown to alter the plaque–gingivitis relationship. With the onset of puberty, both female and male children exhibit significantly increased gingival bleeding (mean whole mouth score and percent of bleeding sites) that reaches a peak after 1–5 years, without any noticeable changes in plaque levels (Mombelli et al. 1989). Pregnancy was one of the first conditions identified as impacting gingivitis (Löe & Silness 1963, Silness & Löe 1964). Experimental gingivitis studies that compared the same women during pregnancy and 6 months postpartum indicate that, without any significant changes in the amount of plaque accumulated during the two phases, there is more swelling, redness, and bleeding during pregnancy (Raber-Durlacher et al. 1994). Besides puberty and gingivitis, the physiologic changes in hormone levels observed during the menstrual cycle have been associated with infrequent and subtle changes in gingivitis expression (Mariotti 1994). The evidence suggests that hormonal variations during the cycle do not affect clinically normal gingiva but do exacerbate existing chronic gingivitis (Holm-Pedersen & Löe 1967, Kovar et al. 1985, Niemi et al. 1986). The effect of the menstrual cycle has not been investigated in the experimental gingivitis model. Despite the significant impact of menopause on both the systemic and oral health of women (Samsioe 1995, Friedlander 2002), there are limited studies addressing the possible effects of the postmenopausal lack of estrogen or of hormone replacement therapy (HRT) on gingival inflammation. In a study on postmenopausal women receiving HRT, gingival bleeding was significantly decreased compared with the control group of postmenopausal women not receiving HRT; however, oral hygiene was also better in the group receiving HRT (Norderyd et al. 1993). The effect of menopause or HRT has not been investigated in the experimental gingivitis model. The significance of menopause as a metabolic factor modulating the clinical expression of plaque-induced gingival inflammation remains unknown. The effect of contraceptive agents on gingivitis expression, although related to changes in sex hormone levels, will be reviewed under environmental factors.

Table 1. Systemic factors shown to modulate gingivitis expression

Factor/condition	Gingival inflammation modulation
Metabolic	
puberty	+
pregnancy	+
diabetes	+
Genetic	
Down's syndrome	+
Environmental	
smoking	–
vitamin C deficiency	+
antibiotics	–
calcium channel blockers	+
corticosteroids	–
cyclosporin	+
NSAIDs	–
phenytoin	+
Other	
immune (PMN) deficiency	+
leukemia	+
HIV/AIDS	+
psychological stress	+

[+]Heightened gingival response to plaque.
– Reduced gingival response to plaque.
NSAIDs: non-steroidal anti-inflammatory drugs.
PMN: polymorphonuclear granulocytes.
HIV: human immunodeficiency virus.
AIDS: acquired immune deficiency syndrome.

Among pathologic changes, diabetes is an endocrine condition with a well-characterized effect on gingivitis. Diabetic subjects, whether affected by the insulin-dependent (de Pommereau et al. 1992) or the non-insulin dependent (Cutler et al. 1999) form of the disease, have significantly higher gingival inflammation compared with non-diabetics with similar plaque levels.

In the course of implementing an experimental gingivitis trial, appropriate subject selection criteria (age, gender, health status) can eliminate the above factors. From a practical point of view, elimination of the menstrual cycle factor may be difficult, if not impossible, if both male and female subjects are expected to participate in the trial.

Genetic factors

As mentioned earlier, the gingival response to plaque may vary significantly between individuals with neither quantitative nor qualitative differences in plaque accumulation (Abbas et al. 1986a, Lie et al. 1995, 1998). Evidence from experimental gingivitis studies indicates genetic conditions can predispose subjects to increased susceptibility to plaque-induced inflammation. One such genetic condition is Down's syndrome (trisomy 21). In the course of experimental gingivitis trials, Down's syndrome subjects manifest more extensive and severe gingival inflammation and much earlier than age- and sex-matched genetically healthy controls, despite no differences in plaque accumulation rates (Reuland-Bosma et al. 1986, 1988). Although the specific gene(s) responsible for the various phenotypic changes observed in Down's syndrome subjects are not known, the genetic basis is unquestionable.

Recent preliminary evidence suggests that specific genetic characteristics may contribute to exacerbated gingival inflammation in response to plaque accumulation. Subjects positive for a specific interleukin-1 (IL-1) genotype exhibit increased susceptibility to experimental gingivitis (Goodson et al. 2000). The report was based on the comparison of seven periodontally healthy IL-1 genotype-positive subjects to 13 periodontally healthy IL-1 genotype-negative subjects. Significant differences in bleeding on probing between the two groups were found after 10 days of comparable plaque accumulation (Goodson et al. 2000). In contrast to these results, another experimental gingivitis report on ten periodontally healthy IL-1 genotype-positive subjects and ten periodontally healthy IL-1 genotype-negative subjects found no differences in gingival inflammation (including bleeding on probing) between the two groups after 21 days of plaque accumulation (Jepsen et al. 2003). Therefore, the significance of IL-1 genotype as a genetic factor modulating the clinical expression of plaque-induced gingival inflammation remains to be established.

The concept of host-dependent variation in gingivitis susceptibility is supported by the results of a published large-scale experimental gingivitis study. In 1979, Wiedemann et al. reported that in a group of 62 subjects, eight did not develop gingivitis within the time of the study (21 days), despite plaque accumulation. Another group of 25 subjects exhibited substantial gingival inflammation within 2 weeks. The remaining 29 subjects formed an intermediate group, which developed gingival inflammation by day 21 (Wiedemann et al. 1979). In a study involving 45 subjects, van der Weijden and colleagues found that 10 subjects consistently exhibited greater than average gingival inflammation, representing a ''susceptible'' group, while six subjects were consistently below average, representing a ''resistant'' group (van der Weijden et al. 1994). The difference in gingivitis susceptibility between the two groups could not be ascribed to qualitative plaque differences (Lie et al. 1995). In accordance with the above, Watts reported that only 18 of 29 subjects were consistent in bleeding from certain sites in response to plaque accumulation (Watts 1978). From the data reported in the above experimental gingivitis studies (Wiedemann et al. 1979, van der Weijden et al. 1994), one can estimate that approximately 13% of subjects represent a ''resistant'' group.

Collectively, the studies reviewed in this section are consistent with the hypothesis of a host-dependent and genetically based modulation of gingivitis expression and provided the motivation for initiating the present series of investigations.

Environmental factors

An environmental factor with well-documented influence on the clinical expression of plaque-induced gingival inflammation is smoking. Smokers, when compared with non-smokers in experimental gingivitis studies, accumulate plaque at the same rate and exhibit significantly less gingival inflammation at similar plaque levels (Bergstrom & Preber 1986, Danielsen et al. 1990, Lie et al. 1998). When examined, no qualitative plaque differences were found between smokers and non-smokers during experimental gingivitis (Lie et al. 1998). It appears that the effect of smoking is mediated, at least in part, by modulation of the local vascular response (Bergstrom et al. 1988). Because of the smoking-induced altered relationship between plaque and gingivitis (Bergstrom & Preber 1986, Danielsen et al. 1990, Lie et al. 1998, Müller et al. 2002), smokers have to be either excluded from experimental gingivitis trials or included as a separate group.

Various medications have been found to have adverse effects involving the gingiva. Several drugs (Seymour 1993), including anti-convulsants such as phenytoin (Angelopoulos 1975), anti-hypertensive calcium channel blockers such as nifedipine (Nery et al. 1995, O'Valle et al. 1995), and the immunosuppressant cyclosporin (Seymour & Jacobs 1992, O'Valle et al. 1995) cause severe gingival enlargement, a reaction related to the plaque-induced gingival inflammation (Seymour et al. 1996). In a way, the above medications can be considered as factors that exacerbate the plaque-induced gingival inflammatory response.

An equally large number of drugs have been shown to curtail plaque-induced gingival inflammation. These include both steroidal (Sutton & Smales 1983, Vogel et al. 1984, Markitziu et al. 1990) and non-steroidal (Vogel et al. 1984, Heasman et al. 1993b, Jones et al. 1999) anti-inflammatory medications, whether systemically administered (Sutton & Smales 1983, Vogel et al. 1984, Markitziu et al. 1990, Heasman et al. 1993b) or topically applied (Vogel et al. 1984, Jones et al. 1999). These agents act by inhibiting the host response, thus inhibiting the clinical expression of gingivitis. Finally, systemic or topical anti-microbial agents inhibit plaque formation and the ensuing gingivitis. Various systemic antibiotics, such as erythromycin (Helovuo & Paunio 1989), metronidazole (Listgarten et al. 1979, Heijl & Lindhe 1980), clindamycin (Heijl & Lindhe 1980), vancomycin (Jensen et al. 1968, Heijl & Lindhe 1980), and tetracyclines (Listgarten et al. 1979, Atilla et al. 1996) have been shown to inhibit plaque accumulation and gingivitis development. Antibiotics such as tetracycline and vancomycin have also been used as a rinse, and have been shown to reduce plaque and inflammation (Löe et al. 1967). The use of an adjunctive anti-microbial product as a rinse [e.g. chlorhexidine (Bosman & Powell 1977, Hefti & Huber 1987, Cappelli et al. 2000), phenolic compounds (Mankodi et al. 1987, Brecx et al. 1990), amine fluoride/stannous fluoride (Hefti & Huber 1987, Brecx et al. 1990, Zimmermann et al. 1993)] or a toothpaste [e.g. triclosan-containing toothpaste (Lindhe et al. 1993)], to complement mechanical oral hygiene procedures has been associated with decreases in plaque and gingival inflammation.

In addition to their direct anti-microbial (anti-plaque) activity, some of these agents appear capable of modulating the host response by altering functions of several cell types involved in inflammatory reactions (Shapira et al. 1997, Yanagihara et al. 1997, Scaglione & Rossoni 1998, Solomon et al. 2000, Kuzin et al. 2001). Therefore, certain anti-microbial agents, whether systemically or locally used, can truly modulate the clinical expression of plaque-induced gingivitis, and not simply reduce gingival inflammation by reducing plaque levels. In this context, it is noteworthy that mouth irrigation as an adjunct to toothbrushing, even if it is performed with just water (Flemmig et al. 1990, Cutler et al. 2000), appears to inhibit plaque-induced inflammation, an effect attributed to reduction in the concentration of inflammatory mediators (such as IL-1β and prostaglandin E_2) in gingival crevicular fluid (Cutler et al. 2000). Obviously, use of any of the above products needs to be excluded in an experimental gingivitis trial aimed at identifying the inherent characteristics of the host response to plaque accumulation.

Although effects similar to the ones associated with changes in sex hormone levels during pregnancy (see section on *Metabolic factors* above) have been reported in earlier studies on young women taking contraceptives (Lindhe et al. 1969, Kalkwarf 1978), recent studies suggest that the effect of newer contraceptive pills (which have much reduced hormone content compared with the ones used in the past) on gingivitis is practically nil (Klinger et al. 1998, Preshaw et al. 2001). Therefore, exclusion of females of childbearing age that use contraceptive medication from experimental gingivitis trials is not supported by current evidence.

Among the various nutritional factors examined during the last 35 years, only ascorbate (vitamin C) has been consistently shown to affect gingivitis expression. In both humans (Leggott et al. 1986, 1991) and non-human primates (Alvares et al. 1981) subclinical deficiency of ascorbate results in increased

gingivitis, relative to non-deficient controls with similar plaque levels and the same type of microflora. Subclinical ascorbate deficiency resulted in 36% increase of mean GI score in non-human primates (Alvares et al. 1981), while in humans it reduced in half the percentage of sites that were healthy (non-bleeding) (Leggott et al. 1986). Elimination of a possible subclinical vitamin C deficiency among participants of an experimental gingivitis trial can be easily accomplished by nutritional supplementation.

Other systemic factors

Other systemic conditions identified as affecting the plaque–gingivitis relationship include neutropenia (Andrews et al. 1965, Rylander et al. 1975, Reichart & Dornow 1978), leukemia (Levin & Kennedy 1973, Bergmann et al. 1992), and human immunodeficiency virus/acquired immune deficiency syndrome (Glick et al. 1990). Psychological stress, at least in adults (Deinzer et al. 1998, 1999, Vanderas et al. 1998), leads to heightened gingivitis expression. However, because psychological (academic) stress results in greater plaque accumulation, the relationship between stress and gingivitis may be mediated, at least in part, by this increase in plaque accumulation (Deinzer et al. 2001). Of the above conditions, stress is the one that is impractical, if not impossible, to eliminate during implementation of an experimental gingivitis trial. However, stress levels can be ascertained through appropriate questionnaires and taken into consideration during data analysis.

Rationale and significance

The susceptibility of an animal host to inflammation in response to various stimuli, including bacteria and bacterial products, varies significantly between strains of the same species (Allen et al. 1983, Yoshino et al. 1985). The physiological and/or genetic basis for the differences among strains has been partly elucidated (Sternberg et al. 1989, Listwak et al. 1999, Kandil et al. 2000), and genetic manipulations, e.g. introduction of a single immune response gene, can significantly alter such host inflammatory responses (Hammer et al. 1990, Tatakis et al. 2002). This biological phenomenon is consistent with the reported variation in plaque-induced gingival inflammation in humans. However, in the case of humans, very limited information is available regarding the physiological/genetic reasons underlying this variation.

It was hypothesized that within a large-scale experimental gingivitis trial, conducted in a manner that minimizes confounding from parameters already known to influence gingivitis expression as described above, it would be possible to identify two subpopulations of individuals with inherently significantly different response to plaque-induced gingival inflammation (Trombelli et al. 2004b). Identification of these two groups of subjects should allow further study of other relevant clinical characteristics, such as their response to therapy (Trombelli et al. 2004a) and characterization of physiologic or genetic parameters that set them apart. Being able to identify physiologic and/or genetic factors that account for increased susceptibility or resistance to the development of plaque-induced inflammation could have implications well beyond gingivitis prevention and management. Such potential far-reaching implications rest upon the unique relationship between gingivitis and periodontitis.

The epidemiology and natural history of gingivitis and periodontitis indicate that gingival inflammation is invariably a component of periodontitis and that gingivitis precedes the onset of periodontitis (Lindhe et al. 1973, Löe et al. 1986). However, it is also incontestable that all gingivitis cases do not proceed to develop into periodontitis (Lindhe et al. 1973, Listgarten et al. 1985, Löe et al. 1986, Prayitno et al. 1993). The reason for this is that plaque bacteria are necessary but not sufficient for the development of periodontitis, a susceptible host is necessary (Lindhe et al. 1973, Page & Schroeder 1982, Löe et al. 1986, Page 1999). The fact that gingivitis is a very poor predictor of periodontitis in persons under the age of 30 years (Prayitno et al. 1993) may also be related to the fact that gingivitis will manifest itself only days or weeks (Löe et al. 1965, Theilade et al. 1966) after the onset of plaque accumulation, while periodontitis is a condition that in the majority of cases requires far longer periods (few years to decades) (Lindhe et al. 1973, Löe et al. 1978) to evolve. At the present time, there are no reliable means to predict susceptibility to periodontitis. What the literature suggests, however, is that susceptibility to periodontitis may be linked to susceptibility to gingivitis.

Studies by van der Velden and colleagues indicate that susceptibility to experimental gingivitis (in terms of development of bleeding on probing) differs between two groups of patients with apparently different susceptibility to periodontitis. The group with greater periodontitis susceptibility exhibited greater susceptibility to gingivitis (van der Velden et al. 1985), a result that could not be explained by the age difference between the two groups (Winkel et al. 1987), or by the specific microbial (plaque) or immune (host) parameters examined in these subjects (Abbas et al. 1986b). The notion of a link between susceptibility to periodontitis and susceptibility to gingivitis is also supported by studies on patients with Down's syndrome. Down's syndrome patients exhibit severe periodontal disease early on (Reuland-Bosma & van Dijk 1986, Modeer et al. 1990), with no evidence of qualitative plaque differences between Down's syndrome patients and controls (Amano et al. 2001), and also manifest greater gingival inflammation much sooner than age- and sex-matched controls despite similar plaque accumulation (Reuland-Bosma et al. 1986, 1988). Therefore, it is possible that identification of factors related to increased susceptibility to gingivitis may help identify, at an early age, subjects at risk for periodontitis. If this were the case, it would permit the targeted application of public health resources and private practice-based preventive measures in a cost-effective manner.

Given the relationship between gingivitis and periodontitis (gingivitis precedes periodontitis, gingivitis is a prerequisite for periodontitis development), efforts to develop effective anti-gingivitis agents remains an important objective in the prevention of periodontitis (Robinson 1995). In the context of such efforts, the development of criteria/tests to identify subjects highly susceptible/resistant to gingivitis may provide a unique and powerful tool for designing and implementing clinical trials aimed at the evaluation of systemic and topical anti-microbial agents and host response modifying agents.

Conclusions

The experimental gingivitis model, initially developed by Löe and coworkers

as a tool to demonstrate the relationship between plaque and gingivitis (Löe et al. 1965, 1967, Theilade et al. 1966), has been repeatedly validated and extensively used in the last 37 years for the study of gingivitis pathogenesis and evaluation of therapeutic interventions, mostly in smaller scale studies. Over the years, several factors that modulate clinical expression of gingivitis have been identified. However, the experimental gingivitis model has never been specifically used in a large-scale trial for the identification of individuals susceptible or resistant to gingivitis development and the subsequent characterization of inherent factors associated with gingivitis susceptibility.

Our choice of this model for the study of genetic and environmental factors modulating susceptibility to gingivitis was based on the following criteria: (a) the model has been extensively validated in humans, non-human primates, and other mammals, (b) the general qualitative and quantitative characteristics of the anticipated responses are well documented and provide an invaluable historical control, and (c) it is the only periodontal disease model that is fully reversible, thus precluding any permanent damage to the participants.

The accompanying papers report: (a) the findings of a large-scale experimental gingivitis trial that was designed and conducted taking into consideration the factors known to modulate the clinical expression of plaque-induced gingivitis, as reviewed here, and the identification of two subpopulations of subjects with significantly different gingival inflammatory response to plaque accumulation (Trombelli et al. 2004b), and (b) the response of these two subpopulations to self-administered plaque control (Trombelli et al. 2004a). Identification of host characteristics associated with resistance or high susceptibility to plaque-induced gingivitis remains the long-term goal of these studies.

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References

Abbas, F., van der Velden, U., Hart, A. A., Moorer, W. R., Vroom, T. M. & Scholte, G. (1986a) Bleeding/plaque ratio and the development of gingival inflammation. *Journal of Clinical Periodontology* **13**, 774–782.

Abbas, F., van der Velden, U., Moorer, W. R., Everts, V., Vroom, T. M. & Scholte, G. (1986b) Experimental gingivitis in relation to susceptibility to periodontal disease. II. Phase-contrast microbiological features and some host–response observations. *Journal of Clinical Periodontology* **13**, 551–557.

Addy, M., Dummer, P. M., Hunter, M. L., Kingdon, A. & Shaw, W. C. (1987) A study of the association of fraenal attachment, lip coverage, and vestibular depth with plaque and gingivitis. *Journal of Periodontology* **58**, 752–757.

Albandar, J. M., Buischi, Y. A. & Axelsson, P. (1995) Caries lesions and dental restorations as predisposing factors in the progression of periodontal diseases in adolescents. A 3-year longitudinal study. *Journal of Periodontology* **66**, 249–254.

Albandar, J. M. & Tinoco, E. M. (2002) Global epidemiology of periodontal diseases in children and young persons. *Periodontology 2000* **29**, 153–176.

Allen, J. B., Calandra, G. B. & Wilder, R. L. (1983) Cutaneous inflammatory reactions to group A streptococcal cell wall fragments in Fisher and Lewis inbred rats. *Infection and Immunity* **42**, 796–801.

Alvares, O., Altman, L. C., Springmeyer, S., Ensign, W. & Jacobson, K. (1981) The effect of subclinical ascorbate deficiency on periodontal health in nonhuman primates. *Journal of Periodontal Research* **16**, 628–636.

Amano, A., Kishima, T., Akiyama, S., Nakagawa, I., Hamada, S. & Morisaki, I. (2001) Relationship of periodontopathic bacteria with early-onset periodontitis in Down's syndrome. *Journal of Periodontology* **72**, 368–373.

Andrews, R. G., Benjamin, S., Shore, N. & Canter, S. (1965) Chronic benign neutropenia of childhood with associated oral manifestations. *Oral Surgery, Oral Medicine, Oral Pathology* **20**, 719–725.

Angelopoulos, A. P. (1975) Diphenylhydantoin gingival hyperplasia. A clinicopathological review. 1. Incidence, clinical features and histopathology. *Journal of the Canadian Dental Association* **41**, 103–106.

Armitage, G. C. (1999) Development of a classification system for periodontal diseases and conditions. *Annals of Periodontology* **4**, 1–6.

Atilla, G., Balcan, M., Bicakci, N. & Kazandi, A. (1996) The effect of non-surgical periodontal and adjunctive minocycline–HCl treatments on the activity of salivary proteases. *Journal of Periodontology* **67**, 1–6.

Bader, J. D., Rozier, R. G., McFall, W. T. Jr. & Ramsey, D. L. (1991) Effect of crown margins on periodontal conditions in regularly attending patients. *Journal of Prosthetic Dentistry* **65**, 75–79.

Bergmann, O. J., Ellegaard, B., Dahl, M. & Ellegaard, J. (1992) Gingival status during chemical plaque control with or without prior mechanical plaque removal in patients with acute myeloid leukaemia. *Journal of Clinical Periodontology* **19**, 169–173.

Bergstrom, J., Persson, L. & Preber, H. (1988) Influence of cigarette smoking on vascular reaction during experimental gingivitis. *Scandinavian Journal of Dental Research* **96**, 34–39.

Bergstrom, J. & Preber, H. (1986) The influence of cigarette smoking on the development of experimental gingivitis. *Journal of Periodontal Research* **21**, 668–676.

Bimstein, E. & Matsson, L. (1999) Growth and development considerations in the diagnosis of gingivitis and periodontitis in children. *Pediatric Dentistry* **21**, 186–191.

Bissada, N. F., Ibrahim, S. I. & Barsoum, W. M. (1974) Gingival response to various types of removable partial dentures. *Journal of Periodontology* **45**, 651–659.

Bosman, C. W. & Powell, R. N. (1977) The reversal of localized experimental gingivitis. A comparison between mechanical toothbrushing procedures and a 0.2% chlorhexidine mouthrinse. *Journal of Clinical Periodontology* **4**, 161–172.

Boyd, R. L. & Baumrind, S. (1992) Periodontal considerations in the use of bonds or bands on molars in adolescents and adults. *Angle Orthodontist* **62**, 117–126.

Brecx, M., Netuschil, L., Reichert, B. & Schreil, G. (1990) Efficacy of listerine, meridol and chlorhexidine mouthrinses on plaque, gingivitis and plaque bacteria vitality. *Journal of Clinical Periodontology* **17**, 292–297.

Brecx, M. C., Lehmann, B., Siegwart, C. M., Gehr, P. & Lang, N. P. (1988) Observations on the initial stages of healing following human experimental gingivitis. A clinical and morphometric study. *Journal of Clinical Periodontology* **15**, 123–129.

Breuer, M. M. & Cosgrove, R. S. (1989) The relationship between gingivitis and plaque levels. *Journal of Periodontology* **60**, 172–175.

Cappelli, D., Holt, S. C., Singer, R. E., Pickrum, H. M. & Ebersole, J. L. (2000) Effects of 0.12% chlorhexidine gluconate on experimental gingivitis in non-human primates: clinical and microbiological alterations. *Oral Diseases* **6**, 124–131.

Chung, C. H., Vanarsdall, R. L., Cavalcanti, E. A., Baldinger, J. S. & Lai, C. H. (2000) Comparison of microbial composition in the subgingival plaque of adult crowded versus non-crowded dental regions. *International Journal of Adult Orthodontics and Orthognathic Surgery* **15**, 321–330.

Ciancio, S. G. (1986) Current status of indices of gingivitis. *Journal of Clinical Periodontology* **13**, 375–378, 381–372.

Ciantar, M. & Caruana, D. J. (1998) Periotron 8000: calibration characteristics and reliability. *Journal of Periodontal Research* **33**, 259–264.

Cutler, C. W., Machen, R. L., Jotwani, R. & Iacopino, A. M. (1999) Heightened gingival inflammation and attachment loss in type 2 diabetics with hyperlipidemia. *Journal of Periodontology* **70**, 1313–1321.

Cutler, C. W., Stanford, T. W., Abraham, C., Cederberg, R. A., Boardman, T. J. & Ross, C. (2000) Clinical benefits of oral irrigation for periodontitis are related to reduction of pro-inflammatory cytokine levels and plaque. *Journal of Clinical Periodontology* **27**, 134–143.

Daneshmand, H. & Wade, A. B. (1976) Correlation between gingival fluid measurements and macroscopic and microscopic characteristics of gingival tissue. *Journal of Periodontal Research* **11**, 35–46.

Danielsen, B., Manji, F., Nagelkerke, N., Fejerskov, O. & Baelum, V. (1990) Effect of cigarette smoking on the transition dynamics in experimental gingivitis. *Journal of Clinical Periodontology* **17**, 159–164.

Deinzer, R., Forster, P., Fuck, L., Herforth, A., Stiller-Winkler, R. & Idel, H. (1999) Increase of crevicular interleukin 1beta under academic stress at experimental gingivitis sites and at sites of perfect oral hygiene. *Journal of Clinical Periodontology* **26**, 1–8.

Deinzer, R., Hilpert, D., Bach, K., Schawacht, M. & Herforth, A. (2001) Effects of academic stress on oral hygiene – a potential link between stress and plaque-associated disease? *Journal of Clinical Periodontology* **28**, 459–464.

Deinzer, R., Ruttermann, S., Mobes, O. & Herforth, A. (1998) Increase in gingival inflammation under academic stress. *Journal of Clinical Periodontology* **25**, 431–433.

de Pommereau, V., Dargent-Pare, C., Robert, J. J. & Brion, M. (1992) Periodontal status in insulin-dependent diabetic adolescents. *Journal of Clinical Periodontology* **19**, 628–632.

Engelberger, T., Hefti, A., Kallenberger, A. & Rateitschak, K. H. (1983) Correlations among papilla bleeding Index, other clinical indices and histologically determined inflammation of gingival papilla. *Journal of Clinical Periodontology* **10**, 579–589.

Fischman, S. L. (1986) Current status of indices of plaque. *Journal of Clinical Periodontology* **13**, 371–374, 379–380.

Flemmig, T. F., Newman, M. G., Doherty, F. M., Grossman, E., Meckel, A. H. & Bakdash, M. B. (1990) Supragingival irrigation with 0.06% chlorhexidine in naturally occurring gingivitis. I. 6 month clinical observations. *Journal of Periodontology* **61**, 112–117.

Friedlander, A. H. (2002) The physiology, medical management and oral implications of menopause. *Journal of the American Dental Association* **133**, 73–81.

Glick, M., Pliskin, M. E. & Weiss, R. C. (1990) The clinical and histologic appearance of HIV-associated gingivitis. *Oral Surgery, Oral Medicine, and Oral Pathology* **69**, 395–398.

Goldstein, A. R. (1979) Enamel pearls as contributing factor in periodontal breakdown. *Journal of the American Dental Association* **99**, 210–211.

Golub, L. M. & Kleinberg, I. (1976) Gingival crevicular fluid: a new diagnostic aid in managing the periodontal patient. *Oral Sciences Reviews* **8**, 49–61.

Goodson, J. M., Palys, M. D. & Socransky, S. S. (2000) Gingival bleeding accentuated by plaque in healthy IL-1(+) genotype subjects. *Journal of Dental Research* **79** (Abstract 221), 171.

Goutoudi, P., Koidis, P. T. & Konstantinidis, A. (1997) Gingival recession: a cross-sectional clinical investigation. *European Journal of Prosthodontics and Restorative Dentistry* **5**, 57–61.

Greene, J. C. & Vermillion, J. R. (1960) The oral hygiene index: a method for classifying oral hygiene status. *Journal of the American Dental Association* **61**, 172–179.

Greenstein, G., Caton, J. & Polson, A. M. (1981) Histologic characteristics associated with bleeding after probing and visual signs of inflammation. *Journal of Periodontology* **52**, 420–425.

Gulati, M. S., Grewal, N. & Kaur, A. (1998) A comparative study of effects of mouth breathing and normal breathing on gingival health in children. *Journal of the Indian Society of Pedodontics and Preventive Dentistry* **16**, 72–83.

Hammer, R. E., Maika, S. D., Richardson, J. A., Tang, J. P. & Taurog, J. D. (1990) Spontaneous inflammatory disease in transgenic rats expressing HLA-B27 and human beta 2m: an animal model of HLA-B27-associated human disorders. *Cell* **63**, 1099–1112.

Hancock, E. B., Cray, R. J. & O'Leary, T. J. (1979) The relationship between gingival crevicular fluid and gingival inflammation. A clinical and histologic study. *Journal of Periodontology* **50**, 13–19.

Hart, T. C. & Kornman, K. S. (1997) Genetic factors in the pathogenesis of periodontitis. *Periodontology 2000* **14**, 202–215.

Heasman, P. A., Collins, J. G. & Offenbacher, S. (1993a) Changes in crevicular fluid levels of interleukin-1 beta, leukotriene B4, prostaglandin E2, thromboxane B2 and tumour necrosis factor alpha in experimental gingivitis in humans. *Journal of Periodontal Research* **28**, 241–247.

Heasman, P. A., Offenbacher, S., Collins, J. G., Edwards, G. & Seymour, R. A. (1993b) Flurbiprofen in the prevention and treatment of experimental gingivitis. *Journal of Clinical Periodontology* **20**, 732–738.

Hefti, A. F. & Huber, B. (1987) The effect on early plaque formation, gingivitis and salivary bacterial counts of mouthwashes containing hexetidine/zinc, aminefluoride/tin or chlorhexidine. *Journal of Clinical Periodontology* **14**, 515–518.

Heijl, L. & Lindhe, J. (1980) Effect of selective antimicrobial therapy on plaque and gingivitis in the dog. *Journal of Clinical Periodontology* **7**, 463–478.

Helovuo, H. & Paunio, K. (1989) Effects of penicillin and erythromycin on the clinical parameters of the periodontium. *Journal of Periodontology* **60**, 467–472.

Herrmann, J. M., Gonzales, J. R., Boedeker, R. H., Vonholdt, J. & Meyle, J. (2001) Micro-assay for the detection of elastase activity in the gingival crevice. *Journal of Clinical Periodontology* **28**, 31–37.

Hinrichs, J. E., Bandt, C. L. & Smith, J. A. (1984a) Relative error (variability) associated with an improved instrument for measuring gingival crevicular fluid. *Journal of Periodontology* **55**, 294–298.

Hinrichs, J. E., Bandt, C. L., Smith, J. A. & Golub, L. M. (1984b) A comparison of 3 systems for quantifying gingival crevicular fluid with respect to linearity and the effects of qualitative differences in fluids. *Journal of Clinical Periodontology* **11**, 652–661.

Holm-Pedersen, P. & Löe, H. (1967) Flow of gingival exudate as related to menstruation and pregnancy. *Journal of Periodontal Research* **2**, 13–20.

Holt, S. C., Ebersole, J., Felton, J., Brunsvold, M. & Kornman, K. S. (1988) Implantation of *Bacteroides gingivalis* in nonhuman primates initiates progression of periodontitis. *Science* **239**, 55–57.

Hou, G. L. & Tsai, C. C. (1993) Relationship between palato-radicular grooves and localized periodontitis. *Journal of Clinical Periodontology* **20**, 678–682.

Hugoson, A. (1971) Gingivitis in pregnant women. A longitudinal clinical study. *Odontologisk Revy* **22**, 65–84.

Jacobson, L. (1973) Mouthbreathing and gingivitis. 1. Gingival conditions in children with epipharyngeal adenoids. *Journal of Periodontal Research* **8**, 269–277.

Jalil, R. A., Cornick, D. E. & Waite, I. M. (1983) Effect of variation in dietary sucrose intake on plaque removal by mechanical means. *Journal of Clinical Periodontology* **10**, 389–398.

Jensen, S. B., Löe, H., Schiott, C. R. & Theliade, E. (1968) Experimental gingivitis in man. 4. Vancomycin induced changes in bacterial plaque composition as related to development of gingival inflammation. *Journal of Periodontal Research* **3**, 284–293.

Jepsen, S., Eberhard, J., Fricke, D., Hedderich, J., Siebert, R. & Acil, Y. (2003) Interleukin-1 gene polymorphisms and experimental gingivitis. *Journal of Clinical Periodontology* **30**, 102–106.

Jones, D. S., Irwin, C. R., Woolfson, A. D., Djokic, J. & Adams, V. (1999) Physicochemical characterization and preliminary in vivo efficacy of bioadhesive, semisolid formulations containing flurbiprofen for the treatment of gingivitis. *Journal of Pharmaceutical Sciences* **88**, 592–598.

Kalkwarf, K. L. (1978) Effect of oral contraceptive therapy on gingival inflammation in humans. *Journal of Periodontology* **49**, 560–563.

Kandil, H. M., Argenzio, R. A. & Sartor, R. B. (2000) Low endogenous prostaglandin E2 predisposes to relapsing inflammation in experimental rat enterocolitis. *Digestive Diseases and Sciences* **45**, 2091–2099.

Klinger, G., Eick, S., Pfister, W., Graser, T., Moore, C. & Oettel, M. (1998) Influence of hormonal contraceptives on microbial flora of gingival sulcus. *Contraception* **57**, 381–384.

Kovar, M., Jany, Z. & Erdelsky, I. (1985) Influence of the menstrual cycle on the gingival microcirculation. *Czechoslovak Medicine* **8**, 98–103.

Kuzin, II, Snyder, J. E., Ugine, G. D., Wu, D., Lee, S., Bushnell, T. Jr., Insel, R. A., Young, F. M. & Bottaro, A. (2001) Tetracyclines inhibit activated B cell function. *International Immunology* **13**, 921–931.

Lamster, I. B., Vogel, R. I., Hartley, L. J., DeGeorge, C. A. & Gordon, J. M. (1985) Lactate dehydrogenase, beta-glucuronidase and arylsulfatase activity in gingival crevicular fluid associated with experimental gingivitis in man. *Journal of Periodontology* **56**, 139–147.

Lang, N. P., Kiel, R. A. & Anderhalden, K. (1983) Clinical and microbiological effects of subgingival restorations with overhanging or clinically perfect margins. *Journal of Clinical Periodontology* **10**, 563–578.

Laskaris, G. & Scully, C. (2003) *Periodontal manifestations of local and systemic diseases: colour atlas and text*. Berlin: Springer.

Leggott, P. J., Robertson, P. B., Jacob, R. A., Zambon, J. J., Walsh, M. & Armitage, G. C. (1991) Effects of ascorbic acid depletion and supplementation on periodontal health and subgingival microflora in humans. *Journal of Dental Research* **70**, 1531–1536.

Leggott, P. J., Robertson, P. B., Rothman, D. L., Murray, P. A. & Jacob, R. A. (1986) The effect of controlled ascorbic acid depletion and supplementation on periodontal health. *Journal of Periodontology* **57**, 480–485.

Levin, S. M. & Kennedy, J. E. (1973) Relationship of plaque and gingivitis in patients with leukemia. *Virginia Dental Journal* **50**, 22–25.

Lie, M. A., Danser, M. M., van der Weijden, G. A., Timmerman, M. F., de Graaff, J. & van der Velden, U. (1995) Oral microbiota in subjects with a weak or strong response in experimental gingivitis. *Journal of Clinical Periodontology* **22**, 642–647.

Lie, M. A., van der Weijden, G. A., Timmerman, M. F., Loos, B. G., van Steenbergen, T. J. & van der Velden, U. (1998) Oral microbiota in smokers and non-smokers in natural and experimentally-induced gingivitis. *Journal of Clinical Periodontology* **25**, 677–686.

Lindhe, J., Attstrom, R. & Bjorn, A. L. (1969) The influence of progestogen on gingival exudation during menstrual cycles. A longitudinal study. *Journal of Periodontal Research* **4**, 97–102.

Lindhe, J., Hamp, S. & Löe, H. (1973) Experimental periodontitis in the beagle dog. *Journal of Periodontal Research* **8**, 1–10.

Lindhe, J., Rosling, B., Socransky, S. S. & Volpe, A. R. (1993) The effect of a triclosan-containing dentifrice on established plaque and gingivitis. *Journal of Clinical Periodontology* **20**, 327–334.

Listgarten, M. A., Lindhe, J. & Parodi, R. (1979) The effect of systemic antimicrobial therapy on plaque and gingivitis in dogs. *Journal of Periodontal Research* **14**, 65–75.

Listgarten, M. A., Schifter, C. C. & Laster, L. (1985) 3-year longitudinal study of the periodontal status of an adult population with gingivitis. *Journal of Clinical Periodontology* **12**, 225–238.

Listwak, S., Barrientos, R. M., Koike, G., Ghosh, S., Gomez, M., Misiewicz, B. & Sternberg, E. M. (1999) Identification of a novel inflammation-protective locus in the Fischer rat. *Mammalian Genome* **10**, 362–365.

Löe, H., Anerud, A., Boysen, H. & Morrison, E. (1986) Natural history of periodontal disease in man. Rapid, moderate and no loss of attachment in Sri Lankan laborers 14 to 46 years of age. *Journal of Clinical Periodontology* **13**, 431–445.

Löe, H., Anerud, A., Boysen, H. & Smith, M. (1978) The natural history of periodontal disease in man. The rate of periodontal destruction before 40 years of age. *Journal of Periodontology* **49**, 607–620.

Löe, H. & Silness, J. (1963) Periodontal disease in pregnancy. I. Prevalence and severity. *Acta Odontologica Scandinavica* **21**, 533–551.

Löe, H., Theilade, E. & Jensen, S. B. (1965) Experimental gingivitis in man. *Journal of Periodontology* **36**, 177–187.

Löe, H., Theilade, E., Jensen, S. B. & Schiott, C. R. (1967) Experimental gingivitis in man. 3. Influence of antibiotics on gingival plaque development. *Journal of Periodontal Research* **2**, 282–289.

Lundy, F. T., Chalk, R., Lamey, P. J., Shaw, C. & Linden, G. J. (2001) Quantitative analysis of MRP-8 in gingival crevicular fluid in periodontal health and disease using microbore HPLC. *Journal of Clinical Periodontology* **28**, 1172–1177.

Mankodi, S., Ross, N. M. & Mostler, K. (1987) Clinical efficacy of listerine in inhibiting and reducing plaque and experimental gingivitis. *Journal of Clinical Periodontology* **14**, 285–288.

Mariotti, A. (1994) Sex steroid hormones and cell dynamics in the periodontium. *Critical Reviews in Oral Biology and Medicine* **5**, 27–53.

Mariotti, A. (1999) Dental plaque-induced gingival diseases. *Annals of Periodontology* **4**, 7–19.

Markitziu, A., Zafiropoulos, G., Flores de Jacoby, L. & Pisanty, S. (1990) Periodontal alterations in patients with pemphigus vulgaris taking steroids. A biannual assessment. *Journal of Clinical Periodontology* **17**, 228–232.

Michalowicz, B. S., Diehl, S. R., Gunsolley, J. C., Sparks, B. S., Brooks, C. N., Koertge, T. E., Califano, J. V., Burmeister, J. A. & Schenkein, H. A. (2000) Evidence of a substantial genetic basis for risk of adult periodontitis. *Journal of Periodontology* **71**, 1699–1707.

Modeer, T., Barr, M. & Dahllof, G. (1990) Periodontal disease in children with Down's syndrome. *Scandinavian Journal of Dental Research* **98**, 228–234.

Mombelli, A., Gusberti, F. A., van Oosten, M. A. & Lang, N. P. (1989) Gingival health and gingivitis development during puberty. A 4-year longitudinal study. *Journal of Clinical Periodontology* **16**, 451–456.

Moore, W. E., Holdeman, L. V., Smibert, R. M., Cato, E. P., Burmeister, J. A., Palcanis, K. G. & Ranney, R. R. (1984) Bacteriology of experimental gingivitis in children. *Infection and Immunity* **46**, 1–6.

Mühlemann, H. R. (1977) Psychological and chemical mediators of gingival health. *Journal of Preventive Dentistry* **4**, 6–17.

Müller, H. P., Heinecke, A. & Eger, T. (2000) Site-specific association between supragingival plaque and bleeding upon probing in young adults. *Clinical Oral Investigations* **4**, 212–218.

Müller, H. P., Stadermann, S. & Heinecke, A. (2002) Longitudinal association between plaque and gingival bleeding in smokers and non-smokers. *Journal of Clinical Periodontology* **29**, 287–294.

Nery, E. B., Edson, R. G., Lee, K. K., Pruthi, V. K. & Watson, J. (1995) Prevalence of nifedipine-induced gingival hyperplasia. *Journal of Periodontology* **66**, 572–578.

Niemi, M. L., Ainamo, J. & Sandholm, L. (1986) The occurrence of gingival brushing lesions during 3 phases of the menstrual cycle. *Journal of Clinical Periodontology* **13**, 27–32.

Norderyd, O. M., Grossi, S. G., Machtei, E. E., Zambon, J. J., Hausmann, E., Dunford, R. G. & Genco, R. J. (1993) Periodontal status of women taking postmenopausal estrogen supplementation. *Journal of Periodontology* **64**, 957–962.

O'Valle, F., Mesa, F., Aneiros, J., Gomez-Morales, M., Lucena, M. A., Ramirez, C., Revelles, F., Moreno, E., Navarro, N. & Caballero, T., Masseroli, M. & Garcia del Moral, R. (1995) Gingival overgrowth induced by nifedipine and cyclosporin A. Clinical and morphometric study with image analysis. *Journal of Clinical Periodontology* **22**, 591–597.

Oliver, R. C., Brown, L. J. & Löe, H. (1998) Periodontal diseases in the United States population. *Journal of Periodontology* **69**, 269–278.

Oliver, R. C., Holm-Pedersen, P. & Löe, H. (1969) The correlation between clinical scoring exudate measurements and microscopic evaluation of inflammation in the gingiva. *Journal of Periodontal Research* **4**, 160.

Page, R. C. (1999) Milestones in periodontal research and the remaining critical issues. *Journal of Periodontal Research* **34**, 331–339.

Page, R. C. & Schroeder, H. E. (1982) *Periodontitis in man and other animals. A comparative review*. New York: Karger.

Patters, M. R., Niekrash, C. E. & Lang, N. P. (1989) Assessment of complement cleavage

in gingival fluid during experimental gingivitis in man. *Journal of Clinical Periodontology* **16**, 33–37.

Persson, G. R., Engel, D., Whitney, C., Darveau, R., Weinberg, A., Brunsvold, M. & Page, R. C. (1994) Immunization against *Porphyromonas gingivalis* inhibits progression of experimental periodontitis in nonhuman primates. *Infection and Immunity* **62**, 1026–1031.

Polson, A. M. (1977) Periodontal destruction associated with vertical root fracture. *Journal of Periodontology* **48**, 27–32.

Prayitno, S. W., Addy, M. & Wade, W. G. (1993) Does gingivitis lead to periodontitis in young adults? *Lancet* **342**, 471–472.

Preshaw, P. M., Knutsen, M. A. & Mariotti, A. (2001) Experimental gingivitis in women using oral contraceptives. *Journal of Dental Research* **80**, 2011–2015.

Quigley, G. A. & Hein, J. W. (1962) Comparative cleansing efficiency of manual and power brushing. *Journal of the American Dental Association* **65**, 26–29.

Raber-Durlacher, J. E., van Steenbergen, T. J., van der Velden, U., de Graaff, J. & Abraham-Inpijn, L. (1994) Experimental gingivitis during pregnancy and post-partum: clinical, endocrinological, and microbiological aspects. *Journal of Clinical Periodontology* **21**, 549–558.

Reichart, P. A. & Dornow, H. (1978) Gingivoperiodontal manifestations in chronic benign neutropenia. *Journal of Clinical Periodontology* **5**, 74–80.

Reuland-Bosma, W., Liem, R. S., Jansen, H. W., van Dijk, L. J. & van der Weele, L. T. (1988) Morphological aspects of the gingiva in children with Down's syndrome during experimental gingivitis. *Journal of Clinical Periodontology* **15**, 293–302.

Reuland-Bosma, W. & van Dijk, J. (1986) Periodontal disease in Down's syndrome: a review. *Journal of Clinical Periodontology* **13**, 64–73.

Reuland-Bosma, W., van Dijk, J. & van der Weele, L. (1986) Experimental gingivitis around deciduous teeth in children with Down's syndrome. *Journal of Clinical Periodontology* **13**, 294–300.

Robinson, P. J. (1995) Gingivitis: a prelude to periodontitis? *Journal of Clinical Dentistry* **6**, 41–45.

Rodriguez-Ferrer, H. J., Strahan, J. D. & Newman, H. N. (1980) Effect of gingival health of removing overhanging margins of interproximal subgingival amalgam restorations. *Journal of Clinical Periodontology* **7**, 457–462.

Rüdin, H. J., Overdiek, H. F. & Rateitschak, K. H. (1970) Correlation between sulcus fluid rate and clinical and histological inflammation of the marginal gingiva. *Helvetica Odontologica Acta* **14**, 21–26.

Rylander, H., Attstrom, R. & Lindhe, J. (1975) Influence of experimental neutropenia in dogs with chronic gingivitis. *Journal of Periodontal Research* **10**, 315–323.

Samsioe, G. (1995) The menopause revisited. *International Journal of Gynaecology and Obstetrics* **51**, 1–13.

Scaglione, F. & Rossoni, G. (1998) Comparative anti-inflammatory effects of roxithromycin, azithromycin and clarithromycin. *Journal of Antimicrobial Chemotherapy* **41** (Suppl. B), 47–50.

Schour, I. & Massler, M. (1947) Gingival disease in postwar Italy (1945). I. Prevalence of gingivitis in various age groups. *Journal of the American Dental Association* **35**, 475–482.

Seymour, R. A. (1993) Drug-induced gingival overgrowth. *Adverse Drug Reactions and Toxicological Reviews* **12**, 215–232.

Seymour, R. A. & Jacobs, D. J. (1992) Cyclosporin and the gingival tissues. *Journal of Clinical Periodontology* **19**, 1–11.

Seymour, R. A., Thomason, J. M. & Ellis, J. S. (1996) The pathogenesis of drug-induced gingival overgrowth. *Journal of Clinical Periodontology* **23**, 165–175.

Shapira, L., Schatzker, Y., Gedalia, I., Borinski, R. & Sela, M. N. (1997) Effect of amine and stannous fluoride on human neutrophil functions in vitro. *Journal of Dental Research* **76**, 1381–1386.

Sheiham, A. & Netuveli, G. S. (2002) Periodontal diseases in Europe. *Periodontology 2000* **29**, 104–121.

Sidi, A. D. & Ashley, F. P. (1984) Influence of frequent sugar intakes on experimental gingivitis. *Journal of Periodontology* **55**, 419–423.

Silness, J. & Löe, H. (1964) Periodontal disease in pregnancy. II. Correlation between oral hygiene and periodontal condition. *Acta Odontologica Scandinavica* **22**, 121–135.

Slots, J. & Hausmann, E. (1979) Longitudinal study of experimentally induced periodontal disease in *Macaca arctoides*: relationship between microflora and alveolar bone loss. *Infection and Immunity* **23**, 260–269.

Smukler, H. & Machtei, E. (1987) Gingival recession and plaque control. *Compendium* **8**, 194–198.

Solomon, A., Rosenblatt, M., Li, D. Q., Liu, Z., Monroy, D., Ji, Z., Lokeshwar, B. L. & Pflugfelder, S. C. (2000) Doxycycline inhibition of interleukin-1 in the corneal epithelium. *Investigative Ophthalmology & Visual Science* **41**, 2544–2557.

Sooriyamoorthy, M. & Gower, D. B. (1989) Hormonal influences on gingival tissue: relationship to periodontal disease. *Journal of Clinical Periodontology* **16**, 201–208.

Sternberg, E. M., Hill, J. M., Chrousos, G. P., Kamilaris, T., Listwak, S. J., Gold, P. W. & Wilder, R. L. (1989) Inflammatory mediator-induced hypothalamic–pituitary–adrenal axis activation is defective in streptococcal cell wall arthritis-susceptible Lewis rats. *Proceedings of the National Academy of Sciences of the United States of America* **86**, 2374–2378.

Sutton, R. B. & Smales, F. C. (1983) Cross-sectional study of the effects of immunosuppressive drugs on chronic periodontal disease in man. *Journal of Clinical Periodontology* **10**, 317–326.

Tatakis, D. N., Guglielmoni, P. & Fletcher, H. M. (2002) Accelerated alveolar bone loss in HLA-b27 transgenic rats: an adult onset condition. *Journal of Rheumatology* **29**, 1244–1251.

Theilade, E., Wright, W. H., Jensen, S. B. & Löe, H. (1966) Experimental gingivitis in man. II. A longitudinal clinical and bacteriological investigation. *Journal of Periodontal Research* **1**, 1–13.

Trombelli, L., Scapoli, C., Orlandini, E., Tosi, M., Bottega, S. & Tatakis, D. N. (2004a) Modulation of clinical expression of plaque-induced gingivitis. III. Response of ''high responders''and ''low responders'' to therapy. *Journal of Clinical Periodontology* doi 10.1111/j.1600-051x.2004.00479.x.

Trombelli, L., Tatakis, D. N., Scapoli, C., Bottega, S., Orlandini, E. & Tosi, M. (2004b) Modulation of clinical expression of plaque-induced gingivitis. II. Identification of ''high responder'' and ''low responder'' subjects. *Journal of Clinical Periodontology* doi 10.1111/j.1600-051x.2004.00478.x.

van der Velden, U., Abbas, F. & Hart, A. A. (1985) Experimental gingivitis in relation to susceptibility to periodontal disease. (I). Clinical observations. *Journal of Clinical Periodontology* **12**, 61–68.

van der Weijden, G. A., Timmerman, M. F., Danser, M. M., Nijboer, A., Saxton, C. A. & van der Velden, U. (1994) Effect of pre-experimental maintenance care duration on the development of gingivitis in a partial mouth experimental gingivitis model. *Journal of Periodontal Research* **29**, 168–173.

Vanderas, A. P., Kavvadia, K. & Papagiannoulis, L. (1998) Urinary catecholamine levels and gingivitis in children. *Journal of Periodontology* **69**, 554–560.

Vogel, R. I., Copper, S. A., Schneider, L. G. & Goteiner, D. (1984) The effects of topical steroidal and systemic nonsteroidal anti- inflammatory drugs on experimental gingivitis in man. *Journal of Periodontology* **55**, 247–251.

Waerhaug, J. (1975) Presence or absence of plaque on subgingival restorations. *Scandinavian Journal of Dental Research* **83**, 193–201.

Wagaiyu, E. G. & Ashley, F. P. (1991) Mouth-breathing, lip seal and upper lip coverage and their relationship with gingival inflammation in 11–14 year-old schoolchildren. *Journal of Clinical Periodontology* **18**, 698–702.

Watts, T. L. (1978) Variability of gingival bleeding in experimental gingivitis trials. *Community Dentistry and Oral Epidemiology* **6**, 253–255.

Wiedemann, W., Lahrsow, J. & Naujoks, R. (1979) The effect of periodontal resistance on experimental gingivitis. *Deutsche Zahnarztliche Zeitschrift* **34**, 6–9.

Williams, R. C. (1990) Periodontal disease. *New England Journal of Medicine* **322**, 373–382.

Winkel, E. G., Abbas, F., van der Velden, U., Vroom, T. M., Scholte, G. & Hart, A. A. (1987) Experimental gingivitis in relation to age in individuals not susceptible to periodontal destruction. *Journal of Clinical Periodontology* **14**, 499–507.

Yanagihara, K., Tomono, K., Sawai, T., Hirakata, Y., Kadota, J., Koga, H., Tashiro, T. & Kohno, S. (1997) Effect of clarithromycin on lymphocytes in chronic respiratory *Pseudomonas aeruginosa* infection. *American Journal of Respiratory and Critical Care Medicine* **155**, 337–342.

Yeung, A. L., Lo, E. C., Chow, T. W. & Clark, R. K. (2000) Oral health status of patients 5–6 years after placement of cobalt–chromium removable partial dentures. *Journal of Oral Rehabilitation* **27**, 183–189.

Yoshino, S., Cromartie, W. J. & Schwab, J. H. (1985) Inflammation induced by bacterial cell wall fragments in the rat air pouch. Comparison of rat strains and measurement of arachidonic acid metabolites. *American Journal of Pathology* **121**, 327–336.

Zimmermann, A., Flores-de-Jacoby, L. & Pan, P. (1993) Gingivitis, plaque accumulation and plaque composition under long-term use of meridol. *Journal of Clinical Periodontology* **20**, 346–351.

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