Human viruses in periodontitis

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Periodontitis affects the majority of adults worldwide (4), but relatively few patients receive adequate treatment for the disease (9). Conventional periodontal therapy includes a stabilization phase and a maintenance phase. Stabilization of the disease is accomplished by periodontal mechanical depuration and the removal of calculus and other biofilm-retentive factors, and may involve adjunctive antimicrobial medication and ⁄ or surgery. The long-term goals in the maintenance stage are to have patients exercise proper plaque control and to commit to professional antimicrobial treatment in order to minimize the likelihood of a clinical relapse.

Current periodontal therapy is successful in combating initial and moderate types of periodontitis, but may show limited efficacy in resolving late-stage disease. Optimal periodontal care is often impeded by the lack of patient cooperation and by affordability issues. Periodontal treatment can entail substantial costs attributed to direct healthcare expenses and to loss of income during the time of professional therapy. A greater understanding of the etiopathogeny of periodontal disease seems to be a prerequisite for the development of preventive and therapeutic strategies that are more efficacious and less burdensome for patients.

The task of determining the periodontopathic importance of suspected disease determinants is hampered by difficulty in identifying the initial stage of periodontitis and in distinguishing between progressive and stable phases of the disease. Differences in case definitions and diagnostic methods also complicate the interpretation of epidemiological findings in periodontal research. Periodontitis typically occurs in otherwise healthy individuals and is statistically associated with various environmental and demographic factors (3). The disease can also be linked to rare immunogenetic defects or be part of systemic diseases that primarily affect nonoral tissues (75). It is not clear if some of the proposed risk factors for periodontal disease reflect true genetic or immunological variations, or merely poor health-seeking behavior related to socioeconomic factors, lifestyles or cultural differences.

Microbiological culture and culture-independent molecular studies have identified more than 1,200 bacterial species (140) and 19,000 phylotypes (91) in the oral cavity. At least 400 bacterial species inhabit subgingival sites (141), but despite the long list of different bacteria in periodontitis, fewer than 20 species are considered to be major periodontal pathogens (175, 185). Healthy periodontal sites harbor a scant microbiota of predominantly gram-positive facultative bacteria, whereas periodontitis lesions contain a large variety of gram-negative anaerobic species (171). The shift in the periodontal microbiota with disease development is the result of a multifaceted interaction of microbial-specific traits, host immune responses and ecosystem-based factors. It is not known why fastidious gram-negative anaerobic bacteria outcompete common oral gram-positive bacteria, and why relatively few suspected periodontopathogens are surging in numbers in periodontitis sites.

Periodontopathogenicity is assessed primarily on the basis of an elevated occurrence of a given bacterial species in advanced periodontitis lesions. However, many anaerobic bacteria benefit from proteinaceous components that are present in the gingival crevice fluid and may merely be secondary invaders of periodontitis sites. Also, the cross-sectional design of most bacteria–periodontitis association studies prevents the pathogenetic importance of specific microorganisms from being firmly established. An important exception to this is *Aggregatibacter actinomycetemcomitans* in localized aggressive (juvenile) periodontitis (184). As expected for a true pathogen, the subgingival counts of *A. actinomycetemcomitans* have, in longitudinal studies, shown a dramatic increase immediately prior to clinical attachment loss and a marked decrease at the time of disease remission (16, 64).

Theories proposed so far to explain the etiopathogeny of periodontitis have not been able to

account for several clinical features of the disease (176). Except for localized aggressive (juvenile) periodontitis, destructive periodontal disease develops with unpredictable attacks on the periodontium, and the infectious events that trigger the progression from gingivitis to periodontitis are virtually unknown. The progressive course of periodontitis typically includes prolonged periods of disease remission interrupted by occasional episodes of clinical relapse, and the underlying biological basis is not understood. It remains an enigma why periodontitis of many subjects affects relatively few teeth despite the omnipresence of periodontopathic bacteria in saliva (207). Also, a pure bacterial cause of periodontitis seems unable to explain why the disease tends to develop in a bilaterally symmetrical pattern around the midline of the mouth (103, 122), and why alveolar bone destruction can advance close to the apex at one tooth, while barely affecting the periodontium of a neighboring tooth sharing the same interproximal space (161, 176).

The uncertainty about the infectious and clinical events of periodontal breakdown has given rise to a number of hypotheses about the etiology of periodontitis. Some researchers suggest that specific infectious agents are key to periodontal breakdown. Others emphasize the importance of host immune factors and / or genetic characteristics in the acquisition of periodontitis. The diverse opinion about the etiology of periodontal diseases has led to a variety of classification systems throughout the history of periodontics (7). Some classification systems reflect the view of 'splitters', who emphasize precise definitions and identify a large number of periodontal disease conditions. Other classification systems are formulated by 'lumpers', who search for broad similarities and propose a relatively small number of different types of periodontal disease. A cumbersome or an overly simplified classification system may have limited utility in a clinical practice setting. Periodontal disease classification, like any other type of classification or cataloging system, is intended to be provisional, in other words, it is expected to be supplanted by more rational schemes as disease definitions improve.

It is assumed that periodontitis debuts in genetically or environmentally predisposed individuals, who are infected with virulent infectious agents and reveal persistent gingival inflammation and distinct immune responses (173). Fitting that concept, various herpesviruses have been associated with severe types of periodontal disease (176). Studies on a viral cause for periodontitis mark a turning point in periodontal research, which until recently was centered almost exclusively on a bacterial etiology. Epstein–Barr virus and cytomegalovirus are the most commonly researched viruses in periodontology, and more than one million herpesvirus genome-copies can be present in a single periodontitis site (179). The abundance of herpesviruses in aggressive periodontitis lesions suggests a role of the virus in the development of the disease.

The herpesviral–bacterial hypothesis of periodontitis proposes that an active herpesvirus infection initiates periodontal tissue breakdown and that host immune responses against the herpesvirus infection are an important component of the etiopathogeny of the disease (179). The herpesvirus infection triggers a release of proinflammatory cytokines that have the potential to activate osteoclasts and matrix metalloproteinases and to impair antibacterial immune mechanisms, causing an up-growth of periodontopathic bacteria (179). Herpesviruses and bacteria in aggregate seem capable of explaining several of the clinical features of periodontitis (176). Subject at risk for progressive periodontitis may be managed more efficiently after the mode of acquisition and means of prevention and treatment of periodontal herpesviruses have been delineated.

This article reviews findings on human viruses in periodontal health and disease, and briefly summarizes pathogenetic features of the herpesviral–bacterial mixed periodontal infection. Recent reviews describe in more detail the interaction between periodontal herpesviruses and periodontopathic bacteria (177, 179), and the pathogenic potential of Epstein–Barr virus and cytomegalovirus in oral (178) and nonoral (119, 151) diseases.

Periodontal diseases in immunocompetent subjects

Contemporary studies of periodontal viruses have employed high-performance polymerase chain reaction (PCR) techniques to determine the frequency of the viral genome (single step and nested end-point detection PCR), to quantify viral genome-copies (real-time PCR), or to indicate active viral multiplication (reverse-transcription PCR). PCR-based studies of periodontal herpesviruses have targeted different genomic regions and used methods of different efficiency to extract the target nucleic acid. Negative PCR results may occur because the virus is indeed absent at the time of periodontal sampling or because of technical aspects of the PCR procedure. It is important to use calibrators and internal controls

in PCR assays to exclude false-negative outcomes (24). A false-positive PCR result, which is a rare event using current PCR procedures that have a high degree of assay precision, may be caused by cross-hybridization of nucleotide sequences shared by two or more different DNAs or by sample contamination in the laboratory. The presence of herpesvirus in the periodontium has also been confirmed using labeled DNA probes (105), flow cytometry (45), immuno-fluorescence staining (5) and culture (19).

Periodontal health and gingivitis

The results from periodontal clinical studies can be difficult to interpret because of a lack of agreement about case definitions. Some researchers use the terms 'healthy' and 'normal' periodontium interchangeably to indicate an absence of attachment loss and gingival inflammation. However, 'healthy' gingiva may also be used to describe sites that reveal attachment loss but demonstrate shallow pocket depth and no current inflammation, and 'normal' periodontium may apply to sites that exhibit no attachment loss but show some degree of inflammation. As herpesviruses may have been involved in previous attachment loss and may be present in even slightly inflamed periodontium, ultra-sensitive PCR techniques may identify herpesviruses in 'unhealthy' control sites. Also, healthy periodontal sites of periodontitis patients may harbor more herpesviruses than healthy periodontal sites of individuals with a generally healthy periodontium (50).

Table 1 describes recent studies on herpesvirus presence in healthy periodontium and in gingivitis. Periodontal health is associated with median genome detection rates of 8% for Epstein–Barr virus and for cytomegalovirus (Table 1). Healthy peri-implant sites have demonstrated an absence of cytomegalovirus (132). The observation of few or no herpesvirus genomes in the healthy periodontium is in accordance with a herpesvirus infection of periodontal inflammatory cells (45). Gingivitis studies reveal a median genome detection rate of 20% for Epstein–Barr virus and 33% for cytomegalovirus (Table 1). Herpesvirus-infected periodontal healthy and gingivitis sites typically harbor the viruses in a nontranscriptional state (156, 204) and in copy-counts of only 1,000–20,000 (161).

Periodontitis

The detection rate of periodontal herpesviruses depends on the type of periodontal lesion studied, the viral identification method employed and ethnic / geographical factors (179). As an etiological agent typically peaks during advancing disease and may only occur at low level or be absent in a disease state of remission, studies of periodontal herpesviruses face the difficult task of identifying disease-active and disease-stable periodontitis. One of the most challenging decisions in periodontal classification is to allocate borderline cases to either aggressive periodontitis or chronic periodontitis. The use of patient age as a major criterion for distinguishing between aggressive periodontitis and chronic periodontitis may not be a reliable indicator of active / aggressive or stable / chronic disease. Also, as scaling and root planing of the teeth of immunocompetent individuals may reduce the level of subgingival herpesviruses to low or undetectable (71, 159, 216), which may persist for an extended period of time (159, 196), studies of periodontal herpesviruses should ideally include individuals with no history of receiving professional periodontal treatment.

Probably because of diagnostic difficulties and a natural fluctuation of periodontal herpesviruses, periodontitis studies have reported a wide variation in the occurrence of herpes simplex virus (13–100%), Epstein–Barr virus (3–89%) and cytomegalovirus (0.3–83%) (Table 2). Most studies found higher levels of Epstein–Barr virus (90, 97, 101) and cytomegalovirus (97, 204) in sites of aggressive / progressive periodontitis than in sites of chronic periodontitis, but some studies describe a similar occurrence (85), or even a lower occurrence (13), of the two viruses in aggressive periodontitis (Table 2).

Aggressive periodontitis

Localized aggressive (juvenile) periodontitis is a distinct disease entity that has served as a model for studying periodontal diseases. Localized aggressive periodontitis debuts at puberty and attachment loss occurs at the approximal surfaces of permanent incisors and first molars. The disease can appear in up to half of the children in an affected family. The presence of subgingival herpesviruses was studied in Afro-Caribbean adolescents with classical localized aggressive periodontitis, with incidental periodontal attachment loss, or with a normal periodontium (115). Cytomegalovirus and *Porphyromonas gingivalis* seemed to act synergistically to influence the risk for both the occurrence and the extent of disease. Localized aggressive periodontitis was associated with cytomegalovirus with an odds ratio of 6.6, and with *P. gingivalis* with an odds ratio of 8.7. The odds of having localized aggressive periodontitis increased

Table 1. Prevalence of subgingival genome-copies of herpesviruses in periodontal health and gingivitis

Virus	Healthy/normal periodontium; percentage of positive samples	Gingivitis; percentage of positive samples	Reference
Herpes simplex virus-1	0%	0%	Grenier et al. (71)
	20%	53%	Imbronito et al. (85)
	0%	No data	Nishiyama et al. (130)
	0%	No data	Saygun et al. (160)
	0%	No data	Contreras et al. (41)
Herpes simplex virus-2	0%	No data	Saygun et al. (160)
Epstein–Barr virus	9%	No data	Dawson et al. (49)
	23%	0%	Grenier et al. (71)
	0%	20%	Imbronito et al. (85)
	7%	No data	Chalabi et al. (29)
	8%	No data	Rotola et al. (156)*
	No data	13%	Saygun et al. (161)
	7%	No data	Sunde et al. (195)
	3%	No data	Moghim et al. (121)
	21%	20%	Wu et al. (217)
	No data	30%	Watanabe et al. (215)
	0%	No data	Klemenc et al. (94)
	9%	No data	Konstantinidis et al. (95)
	No data	19%	Li et al. (101)
	6%	No data	Saygun et al. (160)
	18%	No data	Contreras et al. (41)
Median	**8**%	**20**%	
Cytomegalovirus	0%	Not done	Dawson et al. (49)
	8%	25%	Grenier et al. (71)
	57%	40%	Imbronito et al.(85)
	0%	No data	Combs et al. (37)
	0%	No data	Chalabi et al. (29)
	8%	No data	Rotola et al. (156)*
	13%	No data	Ding et al. (52)
	No data	7%	Saygun et al. (161)
	0%	No data	Sunde et al. (195)
	42%	49%	Wu et al. (217)
	18%	No data	Botero et al. (18)
	32%	No data	Chen et al. (32)
	0%	No data	Klemenc et al. (94)

Table 1. *Continued*

Virus	Healthy / normal periodontium; percentage of positive samples	Gingivitis; percentage of positive samples	Reference
	3%	No data	Tantivanich et al. (200)
	0%	No data	Saygun et al. (160)
	9%	No data	Contreras et al. (41)
Median	**8%**	**33%**	
Herpesvirus-6	0%	No data	Klemenc et al. (94)
	0%	No data	Tantivanich et al. (200)
	0%	No data	Contreras et al. (41)
Herpesvirus-7	62%	No data	Rotola et al. (156)*
	0%	No data	Contreras et al. (41)
Herpesvirus-8	0%	No data	Contreras et al. (41)

*Gingival biopsies were examined.

multiplicatively in individuals with a co-infection of cytomegalovirus and *P. gingivalis* (odds ratio, 51.4), compared with the odds of harboring neither of the two infectious agents (115). Thus, localized aggressive periodontitis in Afro-Caribbean adolescents was strongly associated with cytomegalovirus and *P. gingivalis*, and the markedly higher odds ratio of the cytomegalovirus–*P. gingivalis* combined infection than of the sum of the individual pathogens suggests a pathogenetic synergy between the infectious agents.

The relationship between cytomegalovirus activation and disease-active vs. disease-stable periodontitis sites was studied in 11 patients with localized aggressive (juvenile) periodontitis, who were 10–23 years of age and living in Los Angeles (204). Cytomegalovirus transcription of the major capsid protein, a feature suggestive of viral re-activation, was detected in deep periodontal pockets of all five cytomegalovirus-positive patients with early periodontitis (10–14 years of age), but only in one of three cytomegalovirus-positive patients older than 14 years of age, and not in any cytomegalovirus-infected shallow pockets. Individual patients showed transcription of cytomegalovirus genome in some periodontal sites and no transcription in other sites (204). Cytomegalovirus transcription was detected in all five (45%) periodontitis sites lacking radiographic alveolar crestal lamina dura, a feature consistent with ongoing periodontal breakdown (148), but not in four (36%) periodontitis sites that showed radiographic evidence of a lamina dura (204). Also, herpesvirus-like virions, signifying an active viral infection, were

identified in an electronmicroscopic study of aggressive periodontitis lesions (22, 23). The positive association between an active cytomegalovirus infection and aggressive periodontitis suggests involvement of the virus in the disease, but alone cannot differentiate between the possibilities of an active cytomegalovirus infection causing destructive disease, an active cytomegalovirus infection arising secondarily to the pathological changes of disease-active periodontitis, or a combination of these two possibilities in the mode of a vicious cycle.

Ting et al. (204) hypothesized that a primary cytomegalovirus infection at the time of root formation of permanent incisors and first molars can give rise to a defective periodontium. Viruses infecting odontogenic cells of developing hamster teeth can disrupt normal cell differentiation (66), and an active cytomegalovirus infection can change the morphology of developing teeth (88, 190). Perhaps because of a cytomegalovirus infection early in life, teeth affected by localized aggressive periodontitis often show cemental hypoplasia (15). Profound hormonal changes at the onset of puberty may re-activate a periodontal cytomegalovirus infection, resulting in suppression of antibacterial immune defenses and overgrowth of exogenous-like bacteria, such as specific genotypes of *A. actinomycetemcomitans* (93, 161, 179), a major pathogenic species in the early phases of localized aggressive periodontitis (61, 63, 182). Localized aggressive periodontitis lesions harboring an active cytomegalovirus infection tend to be more heavily infected with *A. actinomycetemcomitans* than sites showing a latent cytomegalovirus infection

Table 2. Prevalence of subgingival genome-copies of herpesviruses in aggressive and chronic periodontitis

Virus	Aggressive periodontitis; percentage of positive samples	Chronic periodontitis; percentage of positive samples	Reference
Herpes simplex virus-1	No data	13%	Grenier et al. (71)
	57%	100%	Bilichodmath et al. (13)
	87%	40%	Imbronito et al.(85)
	No data	16%	Grande et al. (69)
	No data	26%	Nishiyama et al. (130)
	No data	31%	Ling et al. (102)
	78%	No data	Saygun et al. (160)
	No data	21%	Contreras et al. (41)
Median	**78**%	**26**%	
Herpes simplex virus-2	0%	16%	Bilichodmath et al. (13)
	17%	No data	Saygun et al. (160)
	No data	0%	Contreras & Slots (43)
Epstein–Barr virus	No data	28%	Dawson et al. (49)
	No data	3%	Grenier et al. (71)
	29%	79%	Bilichodmath et al. (13)
	33%	47%	Imbronito et al.(85)
	No data	79%	Chalabi et al. (29)
	No data	48%	Grande et al. (69)
	55%	46%	Rotola et al. (156)*
	60%	No data	Saygun et al. (161)
	No data	40%	Sunde et al. (195)
	No data	45%	Imbronito et al. (84)
	No data	61%	Moghim et al. (121)
	No data	38%	Wu et al. (217)
	57%	No data	Watanabe et al. (215)
	No data	37%	Wu et al. (216)
	No data	44%	Klemenc et al. (94)
	No data	55%	Konstantinidis et al. (95)
	89%	46%	Kubar et al. (97)
	58%	23%	Li et al. (101)
	No data	4%	Ling et al. (102)
	72%	No data	Saygun et al. (160)
	No data	81%	Contreras et al. (41)
Median	**58**%	**46**%	
Cytomegalovirus	No data	0.3%	Dawson et al. (39)
	No data	35%	Grenier et al. (71)

Table 2. *Continued*

Virus	Aggressive periodontitis; percentage of positive samples	Chronic periodontitis; percentage of positive samples	Reference
	7%	26%	Bilichodmath et al. (13)
	47%	50%	Imbronito et al.(85)
	No data	59%	Chalabi et al. (29)
	No data	80%	Grande et al. (69)
	44%	No data	Ding et al. (52)
	No data	80%	Botero et al. (19)
	53%	No data	Saygun et al. (161)
	No data	12%	Sunde et al. (195)
	No data	83%	Imbronito et al. (84)
	No data	63%	Wu et al. (217)
	40%	60%	Botero et al. (18)
	7%	No data	Watanabe et al. (215)
	No data	59%	Chen et al. (32)
	No data	3%	Klemenc et al. (94)
	78%	27%	Kubar et al. (97)
	No data	34%	Tantivanich et al. (200)
	No data	52%	Ling et al. (102)
	72%	No data	Saygun et al. (160)
	No data	64%	Contreras et al. (41)
Median	**42**%	**52**%	
Herpesvirus-6	No data	24%	Klemenc et al. (94)
	No data	4%	Tantivanich et al. (200)
	No data	71%	Mardirossian et al. (106)†
	No data	0%	Contreras et al. (41)
Herpesvirus-7	No data	67%	Mardirossian et al. (106)†
	64%	62%	Rotola et al. (156)*
	No data	7%	Contreras et al. (41)
Herpesvirus-8	No data	24%	Mardirossian et al. (106)†
	No data	0%	Contreras et al. (41)

*Gingival biopsies were examined.
†Gingival biopsies of human immunodeficiency virus (HIV)-infected individuals were examined.

(204). The affinity of *A. actinomycetemcomitans* for colonizing cytomegalovirus-infected epithelial cells may partly explain the close association of the organism with the disease (201). Also, *A. actinomycetemcomitans* may be able to inhibit epithelial cell proliferation by means of a cytolethal distending toxin (25). Cytomegalovirus-mediated damage to the peri-odontal tissue constituents, antiviral proinflammatory cytokine responses, and bacteria-induced injury of the epithelium, may allow gingival tissue invasion by *A. actinomycetemcomitans* and breakdown of the periodontal attachment and alveolar bone (34).

Aggressive types of periodontitis that affect the majority of teeth in a dentition also exhibit a close

Table 3. Presence of Epstein–Barr virus type 1 and human cytomegalovirus DNAs in deep pockets of progressive and stable periodontitis sites in 16 patients with aggressive periodontitis*

Infectious agents	32 disease-active periodontitis sites	32 disease-stable periodontitis sites	*P*-value (chi-squared test)
Mean ± SD pocket depth at sample sites	6.8 ± 1.6 mm	5.6 ± 1.3 mm	0.002
Epstein–Barr virus-1	14 (44%)†	4 (13%)	0.01
Cytomegalovirus	19 (59%)	4 (13%)	<0.001
Epstein–Barr virus-1 and cytomegalovirus co-infection	9 (29%)	0 (0%)	0.004
Dialister pneumosintes	20 (63%)	6 (19%)	<0.001
Porphyromonas gingivalis	23 (72%)	12 (38%)	0.01
Dialister pneumosintes and *Porphyromonas gingivalis* co-infection	15 (47%)	0 (0%)	<0.001

*From Kamma et al. (90).
†No. (%) of positive sites.

relationship with herpesviruses. In a study from Turkey of young military recruits with generalized aggressive periodontitis, herpes simplex virus type 1, Epstein–Barr virus and cytomegalovirus were each identified in 73–78% of pooled samples from advanced lesions, but the viruses were virtually absent in the subgingival sites of recruits with a healthy periodontium (160). In a Hopi American–Indian population of 75 adolescents, a single individual was found to have generalized aggressive periodontitis and was the only study subject who demonstrated a periodontal dual infection with Epstein–Barr virus type 1 and cytomegalovirus (170).

Kamma et al. (90) studied 16 subjects from Greece with early onset periodontitis (Table 3). Each patient contributed samples from two disease-active sites with an average pocket depth of 6.8 mm and from two disease-stable sites with an average pocket depth of 5.6 mm. Epstein–Barr virus, cytomegalovirus and Epstein–Barr virus–cytomegalovirus co-habitation were significantly associated with disease-active periodontitis. Periodontal cytomegalovirus exhibited a particularly close association with the presence of *P. gingivalis* (180) and *Dialister pneumosintes* (183). Significant associations were also found among *P. gingivalis*, *D. pneumosintes* and *P. gingivalis*–*D. pneumosintes* co-infection, and disease-active periodontitis. Each periodontitis site that demonstrated Epstein–Barr virus–cytomegalovirus co-infection, and all but one site showing *P. gingivalis*–*D. pneumosintes* dual infection, revealed bleeding upon probing, a clinical indicator of an increased risk of progressive disease (100). Parenthetically, Table 3

illustrates how a sole reliance on periodontal pocket depth, without considering periodontitis disease activity, can result in markedly different rates of herpesvirus detection.

A single advanced periodontitis lesion can yield genome copy-counts as high as 8.3×10^8 for Epstein–Barr virus and 4.6×10^5 for cytomegalovirus (98, 161). Other viruses of the herpesvirus family (41, 107) and various nonherpesviruses (137) can also inhabit advanced periodontitis lesions. Hence, the total viral copy-count may approach the total bacterial counts in some diseased periodontal sites.

Chronic periodontitis

Table 4 shows the detection rate of herpesviruses in biopsies from the gingiva of chronic periodontitis lesions of patients living in Los Angeles. The herpesviruses frequently identified were herpes simplex virus-1 (57%), Epstein–Barr virus type 1 (79%) and cytomegalovirus (86%). Herpesviruses can multiply in gingival tissue (73) and tend to reach higher copy-counts in gingival tissue than in subgingival sites (97). Herpesviruses were detected at a significantly lower frequency in biopsies of normal periodontal sites from the same study patients (Table 4). However, the great majority of chronic periodontitis sites, which have a low probability of disease progression, show a latent rather than an active cytomegalovirus infection (19).

Hochman et al. (78) detected antibodies against Epstein–Barr virus in 32%, and against cytomegalovirus in 71%, of gingival crevice fluid samples from 34 study sites. Antibodies against the herpesviruses

Table 4. Herpesvirus genomes in gingival biopsies from chronic periodontitis and from normal periodontal sites*

Herpesviruses	Periodontitis tissue (14 subjects)	Normal periodontium (11 subjects)	*P*-value (chi-squared test)
Herpes simplex virus-1	8 (57%)†	1 (9%)	0.04
Epstein–Barr virus type 1	11 (79%)	3 (27%)	0.03
Epstein–Barr virus type 2	7 (50%)	0	0.02
Human cytomegalovirus	12 (86%)	2 (18%)	0.003
Human herpesvirus-6	3 (21%)	0	0.31
Human herpesvirus-7	6 (43%)	0	0.04
Human herpesvirus-8‡	4 (29%)	0	0.17
Presence of herpesviruses	**14 (100%)**	**5 (45%)**	**0.007**

*From Contreras et al. (41).
†No. (%) of virus-positive patients.
‡Three patients reported to be human immunodeficiency virus (HIV)-positive.

were predominantly of the immunoglobulin A (IgA) isotype in the gingival crevice fluid and of the immunoglobulin G (IgG) isotype in serum samples (78). These antibody findings suggest a local synthesis by plasma cells rather than passive transudation from the blood, and thus provide further evidence of a close relationship between herpesviruses and periodontal disease.

Epstein–Barr virus and cytomegalovirus genomes include regions with substantial polymorphism (147), and herpesvirus subtypes may differ in pathogenicity (217). Epstein–Barr virus exhibits more genotypic variability than recognized previously, which may help to explain specific disease patterns in different geographic areas (30). The Epstein–Barr virus nuclear antigen 2 (EBNA2) genotype 1 occurs more frequently in periodontitis lesions than the EBNA2 genotype 2 (29, 41, 44, 217). In the study of Wu and colleagues (217), EBNA2 genotype 1 was detected in 45%, and EBNA2 genotype 2 was detected in 20% of Chinese patients with chronic periodontitis; however, EBNA2 genotypes 1 and 2 were associated with chronic periodontitis with odds ratios of 2.0 and 8.2, respectively. Wu et al. (217) also identified cytomegalovirus in 79% of patients with chronic periodontitis; moreover, they found the cytomegalovirus gB-I genotype in 20% and the cytomegalovirus gB-II genotype in 87% of cytomegalovirus-positive subjects with periodontitis, and the cytomegalovirus gB-I genotype in 57–59% and the cytomegalovirus gB-II genotype in 47–49% of infected subjects with gingivitis or a normal periodontium. Co-infection with Epstein–Barr virus type 1 and the cytomegalovirus gB-II genotype was associated with periodontitis with an odds ratio of 28.9, compared to an odds ratio of 11.0 for a co-infection with all genotypes of Epstein–

Barr virus and cytomegalovirus (217). Patients who were dually infected with the Epstein–Barr virus type 1 and the cytomegalovirus gB-II genotype tended to have deeper periodontal pocket depths and increased attachment loss (217).

Periodontitis lesions can also harbor papillomaviruses (80, 106, 137), human immunodeficiency virus (HIV) (31, 109), human T-lymphotropic virus type 1 (189), hepatitis B virus (10), hepatitis C virus (108) and torquetenovirus (157). Linkages have been established between human T-lymphotropic virus type 1 infection and gingivitis (odds ratio 3.8) and periodontitis (odds ratio 10.0) (28), between hepatitis B and C viruses and periodontal disease (59), and between torquetenovirus in gingival biopsies and periodontitis (157). The employment of proficient metagenomic pyrosequencing techniques will undoubtedly lead to the identification of several additional periodontal viruses.

Periodontal abscess

The periodontal abscess is characterized by the accumulation of pus that is limited to the marginal periodontium and can appear at a tooth with minor as well as with severe periodontal breakdown (77). A periodontal abscess may arise from mechanical trauma with a foreign object or as a result of periodontal treatment, or without a known cause (77). The early events in the development of a periodontal abscess include multiplication and tissue invasion of one or more subgingival bacterial species (51). Bacteria typically recovered from periodontal abscesses are *Fusobacterium* spp. (75% of abscesses studied), *Prevotella intermedia / nigrescens* (60%), *P. gingivalis* (51%) and *A. actinomycetemcomitans*

(30%) (87). Epstein–Barr virus was detected in 72%, cytomegalovirus in 67%, and co-infection with the two viruses in 56% of 18 abscesses studied, and the herpesviruses were not identified in healthy periodontium or after treatment of the periodontal abscess (163). Hypermobility was present in 90% of abscessed teeth showing a herpesviral dual infection (163). Epstein–Barr virus has also been linked to extra-oral abscesses in children and young adults (192, 199), and cytomegalovirus has been implicated in periodontal (12, 54) and extra-oral (20, 206) abscesses of HIV-infected individuals. It is suggested that re-activation of a periodontal herpesvirus latent infection impairs the periodontal host defense, which permits bacterial pathogens to enter the gingiva and cause abscess formation.

Periodontal diseases in compromised subjects

HIV-associated periodontitis

Cytomegalovirus infection in neonates and immunocompromised patients (HIV-infected patients and transplant recipients) has a high rate of morbidity (178). HIV-induced immunosuppression facilitates herpesvirus re-activation (150), but active herpesviruses may also activate latent HIV (86). Re-activation of latent periodontal herpesviruses by HIV may start a cascade of tissue-destructive events leading to periodontal breakdown. Periodontitis in HIV-infected patients may resemble periodontitis of nonHIV-infected individuals, or be associated with profuse gingival bleeding or necrotizing gingival tissue (79).

Cytomegalovirus was identified in 81% of HIV-associated periodontitis lesions and in 50% of non-HIV-associated periodontitis lesions, and was the most common herpesvirus identified (40). In HIV-infected individuals, cytomegalovirus has also been implicated in acute periodontitis (54), in periodontal abscess formation (12), in mandbular osteomyelitis (12) and in refractory chronic sinusitis (208). Herpesvirus-like virions were detected electron-microscopically in 56% of gingival tissue from HIV-seropositive patients with necrotizing ulcerative periodontitis (35).

Epstein–Barr virus type 1 was identified more frequently in subgingival sites of HIV-positive patients than in subgingival sites of HIV-negative patients (72 vs. 48%) (69). Epstein–Barr virus type 2, which is frequently found in HIV-infected subjects (164, 218), was detected in 57% of biopsies from HIV-associated periodontitis lesions, but was absent in non-HIV-associated periodontitis biopsies (40). Human herpesvirus-8 was present in periodontitis lesions of 24% of HIV-infected individuals with no clinical signs of Kaposi's sarcoma, but was not recovered from periodontitis sites of non-HIV-infected individuals (107). Herpes simplex virus, Epstein–Barr virus, cytomegalovirus and human herpesvirus-8 genomes occur frequently in the saliva of HIV-infected individuals (14, 65, 69), and have been related to ulcerative oral lesions, widespread gingival and mucosal inflammation, and oral cancer (178). The highly active antiretroviral therapy (HAART) may not significantly reduce the prevalence or the load of herpesviruses in saliva (69, 116). The high rate of occurrence of herpesviruses in periodontitis and in oral mucosal pathosis of HIV-infected patients provides substantial evidence of a pathogenetic role of herpesviruses in these diseases.

Necrotizing ulcerative gingivitis / periodontitis

Necrotizing ulcerative gingivitis / periodontitis affects immunocompromised, malnourished and psychosocially stressed individuals. In Europe and the USA, necrotizing ulcerative gingivitis / periodontitis develops primarily in adolescents and young adults and especially in HIV-infected individuals, and almost never in young children. In developing countries, necrotizing gingivitis may expand considerably beyond the periodontium and give rise to the life-threatening disease termed noma or cancrum oris. Noma affects primarily children and is sometimes preceded by a viral infection, such as herpetic gingivostomatitis or measles (89), or HIV (83), which may impair host defenses against resident viruses and pathogenic bacteria. Necrotizing oral diseases have high mortality rates in developing countries because of a high burden of immune-compromising illnesses and limited access to professional diagnosis and treatment.

Contreras et al. (39) studied necrotizing ulcerative gingivitis in nonHIV-infected malnourished Nigerian children, 3–14 years of age (39). Necrotizing gingivitis lesions of malnourished children yielded herpes simplex virus (23% of study lesions), Epstein–Barr virus (27%) and cytomegalovirus (59%), whereas periodontal sites of malnourished, but periodontally normal children revealed virtually no herpesviruses (Table 5). Cytomegalovirus has demonstrated a necrotizing potential in acute retinal necrosis of severely immunocompromised individuals, acute necrotizing esophagitis, necrotizing enterocolitis of preterm

Table 5. Presence of Epstein–Barr virus type 1 and human cytomegalovirus DNAs in acute necrotizing ulcerative gingivitis (ANUG) sites and in normal periodontal sites of malnourished Nigerian children*

Herpesviruses	ANUG + malnutrition (22 subjects)	Normal oral health + malnutrition (20 subjects)	*P*-value (chi-squared test)
Epstein–Barr virus-1	6 (27%)†	1 (5%)	0.13
Cytomegalovirus	13 (59%)	0 (0%)	<0.001
Epstein–Barr virus-1 and cytomegalovirus co-infection	8 (36%)	0 (0%)	0.009

*From Contreras et al. (39).
†No. (%) of virus-positive samples.

infants, necrotizing glomerulonephritis of renal transplant recipients, necrotizing myelitis and necrotizing encephalitis (178). Necrotizing ulcerative gingivitis in African children may be caused by the acquisition of herpesviruses in early childhood (2, 138), impaired immune defenses as a result of malnutrition (57, 128) and an abundance of virulent periodontal bacteria (58, 142).

Acute infection with herpes zoster virus can resemble aspects of necrotizing ulcerative gingivitis and noma, and give rise to osteonecrosis and spontaneous exfoliation of teeth in the region innervated by the affected trigeminal nerve (6, 111). Infectious agents also participate in the type of osteonecrosis of the jaws that is caused by bisphosphonate use for the treatment of osteoporosis, bone complications of cancer, malignant hypercalcaemia and Paget's disease (165). A dental infection can induce necrotizing fasciitis with extensive necrosis in subcutaneous tissues and fascia, and with a high mortality rate (48). The extent to which cytomegalovirus or other herpesviruses are involved in necrotizing diseases of the oral cavity constitutes an important topic for research.

Syndromes

Table 6 lists syndromes that have been associated with both periodontal herpesviruses and severe periodontitis. Medically compromised patients may experience repeated and prolonged herpesvirus re-activation, which may be an important reason for the observed advanced types of periodontitis.

The Guillain–Barré syndrome is an acute neuropathy with limb weakness and with a worldwide annual incidence of 1.3 per 100,000 individuals (99). The syndrome is considered to be an autoimmune disease triggered by a preceding viral or bacterial infection, with the suspected pathogens being Epstein–Barr virus, cytomegalovirus, *Campylobacter jejuni* and *Mycoplasma pneumoniae* (99). Infections may contribute to autoimmunity by converting innate immune responses to adaptive immunity. Periodontitis lesions in a patient with Guillain–Barré syndrome were found to contain cytomegalovirus, but not several other study viruses (198).

The Kostmann syndrome (severe congenital neutropenia) is an autosomal-recessive disease that is characterized by a maturation arrest of neutrophil

Table 6. Human herpesviruses in periodontitis associated with syndromes

Disease	Periodontal viruses	Periodontal disease	References
Guillain–Barré syndrome	Cytomegalovirus	A 37-year-old patient with localized periodontitis	Tabanella & Nowzari (198)
Kostmann syndrome	Epstein–Barr virus	Two siblings (3 and 6 years of age) with severe gingivitis and periodontitis	Yildirim et al. (219)
Fanconi's anemia	Herpes simplex virus, cytomegalovirus	An 11-year-old boy with severe gingivitis and moderate or advanced periodontitis sites	Nowzari et al. (133)
Papillon–Lefèvre syndrome	Epstein–Barr virus, cytomegalovirus	An 11-year-old girl with severe periodontitis around several teeth	Velazco et al. (211)
Down syndrome	Herpes simplex virus (26%), Epstein–Barr virus type 1 (37%), cytomegalovirus (37%)	19 Down syndrome patients with moderate or advanced periodontitis	Hanookai et al. (74)

precursors and extremely low levels of mature peripheral neutrophils (72, 219). The syndrome is associated with recurrent bacterial infections, including cellulitis, perirectal abscess, stomatitis, meningitis, pneumonia, sepsis and severe periodontitis in the primary and permanent dentition (27, 72, 219). Advanced periodontitis lesions in an adult with Kostmann syndrome harbored high proportions of *A. actinomycetemcomitans* (27). Two siblings, 3 and 6 years of age, with the Kostmann syndrome and periodontal disease showed high counts of Epstein–Barr virus in supragingival plaque and in saliva (219).

The Papillon–Lefèvre syndrome is an autosomal-recessive palmoplantar keratodermal disorder, which is associated with severe periodontitis affecting both the primary and the permanent dentition (75). Papillon–Lefèvre periodontitis in the permanent dentition can resemble localized aggressive (juvenile) periodontitis. An 11-year-old girl with Papillon–Lefèvre syndrome harbored Epstein–Barr virus, cytomegalovirus, *A. actinomycetemcomitans*, *P. gingivalis* and other periodontopathic bacteria in deep periodontal lesions (211). The observation that natural killer cell cytotoxicity was depressed by as much as 32–53% compared with control values in 20 patients with Papillon–Lefèvre syndrome may constitute an important pathogenetic aspect of the disease (104). Natural killer cells play a crucial role in the anti-herpesviral host defense (118), and the suppressed killer cell activity may have been induced by cytomegalovirus as an immunoevasive strategy (145).

Fanconi's anemia is an autosomal-recessive disease, which is caused by defective DNA repair, resulting in increased chromosomal instability yielding congenital abnormalities, bone marrow failure and a predisposition to squamous cell carcinomas in the head and neck, and in the anogenital regions (210). Patients with Fanconi's anemia also exhibit increased susceptibility to head-and-neck infections (62, 68) and to periodontitis (158). Periodontitis lesions of an 11-year-old boy with Fanconi's anemia were found to contain a dual infection with herpes simplex virus and cytomegalovirus (133).

Down syndrome is associated with reduced neutrophil and monocyte chemotaxis, impaired neutrophil phagocytosis, reduced T-lymphocyte counts, immature T-lymphocytes and increased susceptibility to respiratory infections, lymphatic leukemia and periodontal disease (74). Down syndrome children have a higher prevalence of seropositivity to herpes simplex virus, Epstein–Barr virus and cytomegalovirus than children without Down syndrome (53, 110). Down syndrome children have also been demonstrated to have an increased oral shedding of cytomegalovirus (53). Hanookai et al. (74) identified herpes simplex virus (26% of study lesions), Epstein–Barr virus (37%) and cytomegalovirus (37%) in periodontitis lesions of Down syndrome patients. Periodontal scaling of the teeth of Down syndrome patients may not ensure a long-term decrease in the amount of subgingival herpesvirus present, possibly in part because of a compromised host defense (74).

Herpesviral–bacterial interactions in periodontitis

A periodontal herpesvirus infection is typically associated with an increased occurrence of periodontopathic bacteria (179). A study of adults with gingivitis or periodontitis found statistically significant associations between periodontal Epstein–Barr virus type 1 or cytomegalovirus and the pathogens *P. gingivalis*, *Tannerella forsythia*, *P. intermedia*, *P. nigrescens* and *Treponema denticola* (44). Quantitative PCR studies of severe periodontitis have revealed a close relationship between genome copy-counts of Epstein–Barr virus and cytomegalovirus and counts of *P. gingivalis* and *T. forsythia* (161). As discussed earlier, cytomegalovirus-infected localized aggressive periodontitis lesions exhibit an elevated occurrence of *P. gingivalis* (115) or *A. actinomycetemcomitans* (204). Similarly, respiratory tract infections, otitis media and other nonoral diseases that were previously thought to be caused solely by bacteria may actually have a combined viral–bacterial etiology (179). Close links among herpesviruses, bacteria and periodontitis are consistent with a role of both types of infectious agents in the pathogenesis of destructive periodontal disease.

Herpesvirus periodontopathic potential

Herpesvirus pathogenicity is complex and is executed through direct virus infection and replication, or via a virally induced alteration of the host immune defense. The early phases of periodontitis in immunologically naïve hosts may predominantly involve cytopathogenic events, whereas most clinical manifestations in immunocompetent individuals are secondary to cellular or humoral immune responses.

Herpesviruses can exert direct cytopathic effects on fibroblasts, keratinocytes, endothelial cells and inflammatory cells, including polymorphonuclear leukocytes, lymphocytes, macrophages and possibly bone cells (42). Epstein–Barr virus and cytomegalovirus can also infect and alter the functions of monocytes, macrophages and lymphocytes in periodontitis lesions (45). Perhaps as result of a herpesvirus periodontal infection, aggressive periodontitis lesions contain fewer overall viable cells, more T-suppressor lymphocytes and more B-lymphocytes (Epstein–Barr virus effect) than chronic periodontitis lesions or healthy periodontal sites (168).

A periodontal herpesvirus infection may increase the pathogenicity of the periodontal microbiota. Herpesvirus proteins expressed on eukaryotic cell membranes may act as new bacterial binding sites (42). Cytomegalovirus can enhance the adherence of *A. actinomycetemcomitans* to primary periodontal pocket epithelial cells and to HeLa cells (201). Herpesviruses may induce abnormalities in the adherence, chemotaxis, phagocytic and bactericidal activities of polymorphonuclear leukocytes (1), which are cells of key importance for the control of periodontopathic bacteria (209). Epstein–Barr virus active infection can also generate anti-neutrophilic antibodies and neutropenia, and polyclonally stimulate the proliferation and differentiation of B-lymphocytes (42). The pathogenic mechanisms of herpesviruses cooperate in exacerbating disease, and probably for that reason, a periodontal dual infection with cytomegalovirus and Epstein–Barr virus (90), or with cytomegalovirus and herpes simplex virus (102), tends to occur in severe types of periodontitis.

The interaction between herpesviruses and bacteria is probably bidirectional, with bacterial enzymes or other inflammation-inducing factors having the potential to activate periodontal herpesviruses (179). Experimental mice infected with murine cytomegalovirus–*P. gingivalis* exhibited a significantly higher mortality rate than mice infected with murine cytomegalovirus–*Escherichia coli* (193). The potential of *P. gingivalis* to suppress the interferon-gamma antiviral host response may partly explain the increase in cytomegalovirus pathogenicity (193).

Antigens of viruses and bacteria play a causal, or at least a contributory, role in destructive periodontal disease. Several studies have shown that T-cell-driven immune responses are activated in patients with periodontitis (82). Specific lymphocyte responses are driven by the nature of the initial antigenic stimuli and are supported by a complex cascade of events involving cytokines, chemokines and other inflammatory mediators (67). Proinflammatory and anti-inflammatory balances controlled by different subsets of lymphocytes are thought to be crucial in the pathogenesis of periodontitis (67).

Epstein–Barr virus and cytomegalovirus infections up-regulate the interleukin-1beta and tumor necrosis factor-alpha gene expression of monocytes and macrophages (42). Increased levels of proinflammatory cytokines in periodontal sites are associated with an enhanced risk of periodontal tissue destruction (134). The herpesvirus-associated proinflammatory cytokines and chemokines can hamper the antibacterial host defense, stimulate bone-resorbing osteoclasts, up-regulate matrix metalloproteinase and down-regulate tissue inhibitors of metalloproteinase, thereby impeding tissue turnover and repair and increasing the risk of periodontal tissue breakdown (120, 134). Also, periodontitis tends to be of greater severity in carriers of the HLA-DR4 alloantigen (127), perhaps because cytomegalovirus-specific CD8^{+} T cells can cross-recognize HLA-DR4 molecules and potentially induce autoimmune reactions (152).

Linkage between periodontitis and nondental diseases

Nonoral diseases

Periodontitis has been associated with increased morbidity and mortality of cardiovascular disease, stroke, pregnancy complications and other medical illnesses (11, 123, 149). Cross-sectional, case-control and cohort studies indicate that periodontitis is linked with a twofold increased risk of cardiovascular disease and with up to a sevenfold higher risk of premature birth (123). Increased periodontal pocket depth has been linked with an odds ratio as high as 8.5 to transient ischemic attack and mild to moderate stroke in an Indian population (146). However, determining the role of periodontal infections in nonoral diseases is fraught with difficulties, and the nature and strength of the relationship between periodontitis and nonoral diseases have been topics of debate (125, 135, 172).

Atherosclerosis is the major contributory factor in most cases of coronary heart disease and ischemic stroke (143). Atherosclerosis may develop as a result of infection with cytomegalovirus, herpes simplex virus, *Chlamydia pneumoniae*, *Helicobacter pylori* or periodontopathic bacteria (124). Cytomegalovirus seropositivity has been linked to cardiovascular disease, defined as a history of stroke, heart attack

and / or congestive heart failure, even after adjusting for confounders (169). Cytomegalovirus shows a particularly close relationship to re-stenosis following coronary angioplasty (21), to myocardial infarction in organ transplant recipients (21) and to abdominal aortic aneurysm (70). Cytomegalovirus and herpes simplex virus, especially when acting together, have the potential to promote the inflammatory and procoagulant environment that underlies the pathogenesis of atherosclerosis (212). The pro-atherogenic potential of cytomegalovirus is based upon changes in the functionalities of smooth muscle cells, macrophages and endothelial cells (188). Cytomegalovirus infection of vascular cells induces cell activation, which leads to the expression of adhesion proteins, major histocompatability complex molecules and cytokine receptors, and to the release of cytokines and growth factors (187). A cytomegalovirus infection in murine models induced high levels of proinflammatory cytokines as well as inflammation of the endocardium, epicardium and myocardium (153) and was a risk factor for increased arterial blood pressure and a co-factor in aortic atherosclerosis (33). Herpes simplex virus and periodontopathic bacteria in aggregate (213) and herpesvirus-8 (46) have also been associated with vascular disease. Conceivably, cytomegalovirus and herpes simplex virus may participate in the pathogenesis of both periodontitis and vascular diseases. This raises the question of whether periodontitis *per se* plays a causal role in vascular diseases, or if some cases of periodontitis and vascular disease develop independently of each other as separate herpesvirus infections.

A link between periodontitis and spontaneous premature birth has been proposed in several, but not in all, studies (113). Initial data have also associated periodontitis with late miscarriage (60) and pre-eclampsia (38). Women of low socioeconomic position, especially African–American women (81), are at elevated risk of developing periodontitis (17) and of experiencing premature birth (166). Again, cytomegalovirus may in some patients be a common etiology of periodontitis and premature birth. US children and adults with a low family income and low level of education, and of nonwhite race / ethnicity, are most likely to be infected with cytomegalovirus and herpes simplex virus (55, 56), and the householder nativity is an important determinant of cytomegalovirus transmission among pre-adolescents (191). Cytomegalovirus can be transmitted transplacentally to the fetus, and a proportion of premature births are caused by a congenital cytomegalovirus infection (8), which occurs with an overall birth

prevalence rate of 0.64% (92). Fetal transmission of cytomegalovirus takes place in 30–60% of women acquiring a primary cytomegalovirus infection during pregnancy and in 0.5–2% of women with recurrent cytomegalovirus infection, in spite of preconception immunity (129). More children in the USA are affected with a congenital cytomegalovirus infection (8,000 annually) than with better known childhood conditions, such as Down syndrome, fetal alcohol syndrome or spina bifida (154), but relatively few women know about congenital cytomegalovirus illness (155). About 11% of live-born infants with congenital cytomegalovirus infection develop permanent disabilities, including hearing loss, vision loss, motor impairments and mental retardation (92). Cytomegalovirus infection transmitted via human milk is mild, self-limiting and without sequelae (26), but gives rise to a permanent infection with the virus (126). Means of preventing primary cytomegalovirus infection before and during pregnancy include frequent handwashing, not sharing drinking glasses or eating utensils with young children (who are most likely to have an active cytomegalovirus infection) and not kissing young children on the mouth (155). Antiviral drugs and hyperimmune globulin therapy may be used in pregnant women with a suspected primary cytomegalovirus infection (36). Of interest, a vaccine (based upon recombinant cytomegalovirus glycoprotein B subunit antigen) to prevent cytomegalovirus infection in seronegative women of childbearing age has shown 50% efficacy (139).

Periodontitis has been statistically associated with several other diseases / conditions, including rheumatoid arthritis (144), which in turn has been linked to Epstein–Barr virus (112, 205) and cytomegalovirus (112); with a variety of renal diseases (96), which have been associated with cytomegalovirus and several other viruses (214); and with premature death from neoplasms and from vascular and digestive diseases (186), which can have a herpesviral etiology (178). A herpesvirus infection affecting different sites in the body may, in some patients, be responsible for the perceived link between periodontitis and these medical illnesses.

P. gingivalis has frequently been related to nonoral diseases (167). The *P. gingivalis*–medical disease link may, at least in part, be the consequence of a periodontal herpesvirus infection which is capable of stimulating an overgrowth of periodontal *P. gingivalis* (180). Also, elevated serum levels of C-reactive protein are associated with aggressive periodontitis (136, 194) and with various nonoral diseases (47). A herpesvirus infection may induce high levels of

C-reactive protein in both periodontal and medical diseases (117).

It thus appears that the biological link between periodontitis and some cases of medical illness may be a shared herpesvirus infection and herpesvirus-induced molecular pathways. If so, conventional periodontal therapy directed at bacteria may not prevent systemic diseases that involve herpesviruses. Periodontal treatment of pregnant women in a large multicenter study caused a significant reduction in the number of periodontopathic bacteria (131), but was unable to alter the rate of preterm birth, low birth weight or fetal growth restriction (114). However, as periodontitis lesions may constitute a major site of herpesvirus multiplication and introduction of active (infectious) herpesviruses into the bloodstream, periodontal treatment employing both antiviral agents and antibacterial debridement may be more effective in preventing systemic dissemination of pathogenic agents and possibly nonoral illnesses. Povidone-iodine and sodium hypochlorite solutions applied subgingivally are potent antiviral and antimicrobial chemotherapeutics (174). Systemic valacyclovir can result in long-term suppression of Epstein-Barr virus and probably also of other herpesviruses in subgingival sites (196), and within gingival tissue harboring high viral loads (97). No data are available on the potential of periodontal treatments targeting both herpesviruses and bacteria to reduce the incidence or the severity of systemic diseases.

Oral diseases

In the oral cavity, periodontitis has been associated with papillomavirus-16-related squamous cell carcinoma of the tongue (202, 203). Co-infection with papillomavirus-18 and Epstein–Barr virus has also been linked to tongue carcinoma (76). As periodontitis lesions frequently harbor papillomaviruses (137), and may even comprise the major oral reservoir of the virus (80), periodontitis sites in intimate contact with the tongue may serve as the source of oncogenic papillomaviruses. A similar hypothesis proposes that periodontitis lesions supply Epstein–Barr virus for the development of hairy leukoplakia of the tongue (181). Papillomaviruses have also been associated with the potentially malignant disorders of oral leukoplakia and oral lichen planus (197). Cytomegalovirus may play a role in the development of the non-neoplastic peripheral giant cell granuloma around teeth (162). Studies are needed to determine if the treatment of periodontal viral infections can decrease the incidence of oral tumors.

Conclusions

A solid understanding of the etiology of periodontitis is critical for developing clinically relevant classification systems and therapies that can ensure long-lasting disease control. Research during the past 15 years has implied that herpesviruses are involved in the etiopathogeny of destructive periodontal disease. It appears that a high periodontal load of active Epstein–Barr virus or cytomegalovirus is statistically associated with aggressive periodontitis, and latent herpesvirus infections are preferentially found in chronic periodontitis and gingivitis sites. Co-infection with Epstein–Barr virus and cytomegalovirus shows a particularly close link with progressive periodontitis. Also, specific genotypes of herpesvirus species may exhibit increased periodontopathic potential. Because of the high copy-counts of Epstein–Barr virus and cytomegalovirus in aggressive periodontitis, it is unlikely that these pathogenic viruses are acting merely as innocuous bystanders present in proportion to the severity of the underlying periodontal pathosis. However, herpesviruses are probably not stand-alone periodontopathic agents, but cooperate with specific bacteria in periodontal tissue breakdown. A co-infection of active herpesviruses and periodontopathic bacteria may constitute a major cause of periodontitis and explain a number of the clinical characteristics of the disease. Conversely, it is implied that herpesvirus negativity is an indicator of a favorable periodontal prognosis. Papillomaviruses and other mammalian viruses are also frequent inhabitants of periodontitis lesions, but their role, if any, in the pathogenesis of the disease is unknown.

The ability of an active herpesvirus infection to alter the periodontal immune responses may constitute a crucial pathogenetic feature of periodontitis. An active herpesvirus infection can exert direct cytopathogenic effects on key cells of the periodontium, induce the release of proinflammatory cytokines with the potential to activate osteoclasts and matrix metalloproteinases, impair host defense mechanisms to create a milieu for the up-growth of periodontopathic bacteria, or give rise to a combination of these pathogenetic events. Repeated and long-standing activation of periodontal herpesviruses, as occurring in immunologically naïve and in immunosuppressed individuals, may increase the frequency and severity of clinical exacerbation of periodontal disease.

The current paradigm of the pathogenesis of periodontitis needs to be revisited based upon the

concept of a herpesviral–bacterial co-infection. Periodontitis may develop stepwise in a series of simultaneous or sequential infectious disease events, including (i) a high herpesvirus load (gingivitis level) in periodontal sites, (ii) activation of periodontal herpesviruses, (iii) an insufficient antiviral cytotoxic T-lymphocyte response, (iv) the presence of specific periodontal pathogenic bacteria, and (v) an inadequate antibacterial antibody response. In most individuals, these five suggested pathogenic determinants of periodontitis may collaborate in a detrimental constellation relatively infrequently and mainly during periods of suppressed cellular immunity. Herpesviruses play a major role as activators of the disease process in this model of periodontitis. Indeed, herpesviruses may be a key missing piece of the periodontopathogenetic jigsaw puzzle that would explain the transition from gingivitis to periodontitis or from stable to progressive periodontitis. Herpesvirus infections of both periodontal and nonoral sites may also be responsible for some of the relationships observed between periodontitis and various medical diseases.

Ongoing research on herpesvirus infections of the periodontium may produce significant progress in the prevention and treatment of periodontitis. Viral studies may lead to clarification of the clinical and biological features of periodontitis, and to new strategies for managing the disease. Detection or quantification of periodontal herpesviruses may prove to have prognostic significance. Assessment of the re-activation status of a periodontal herpesvirus infection may help to guide the treatment of patients with severe periodontitis. Development of anti-herpesvirus vaccines in the not-too-distant future offers real hope for low-cost prevention of periodontitis in large groups of individuals. The existing information justifies adding human periodontitis to the list of diseases that has Epstein–Barr virus, cytomegalovirus and perhaps other human viruses as likely contributory causes.

References

1. Abramson JS, Mills EL. Depression of neutrophil function induced by viruses and its role in secondary microbial infections. *Rev Infect Dis* 1988: **10**: 326–341.
2. Adjei AA, Armah HB, Gbagbo F, Boamah I, Adu-Gyamfi C, Asare I. Seroprevalence of HHV-8, CMV, and EBV among the general population in Ghana, West Africa. *BMC Infect Dis* 2008: **8**: 111.
3. Albander JM. Global risk factors and risk indicators for periodontal diseases. *Periodontol 2000* 2002: **29**: 177–206.
4. Albandar JM. Epidemiology and risk factors of periodontal diseases. *Dent Clin North Am* 2005: **49**: 517–532. v–vi.
5. Amit R, Morag A, Ravid Z, Hochman N, Ehrlich J, Zakay-Rones Z. Detection of herpes simplex virus in gingival tissue. *J Periodontol* 1992: **63**: 502–506.
6. Arikawa J, Mizushima J, Higaki Y, Hoshino J, Kawashima M. Mandibular alveolar bone necrosis after trigeminal herpes zoster. *Int J Dermatol* 2004: **43**: 136–137.
7. Armitage GC. Classifying periodontal diseases – a long-standing dilemma. *Periodontol 2000* 2002: **30**: 9–23.
8. Arvin AM, Fast P, Myers M, Plotkin S, Rabinovich R, National Vaccine Advisory Committee. Vaccine development to prevent cytomegalovirus disease: report from the National Vaccine Advisory Committee. *Clin Infect Dis* 2004: **39**: 233–239.
9. Baelum V, van Palenstein Helderman W, Hugoson A, Yee R, Fejerskov O. A global perspective on changes in the burden of caries and periodontitis: implications for dentistry. *J Oral Rehabil* 2007: **34**: 872–906.
10. Bass BD, Andors L, Pierri LK, Pollock JJ. Quantitation of hepatitis B viral markers in a dental school population. *J Am Dent Assoc* 1982: **104**: 629–632.
11. Beck JD, Offenbacher S. Systemic effects of periodontitis: epidemiology of periodontal disease and cardiovascular disease. *J Periodontol* 2005: **76** (11 Suppl.): 2089–2100.
12. Berman S, Jensen J. Cytomegalovirus-induced osteomyelitis in a patient with the acquired immunodeficiency syndrome. *South Med J* 1990: **83**: 1231–1232.
13. Bilichodmath S, Mangalekar SB, Sharma DC, Prabhakar AK, Reddy SB, Kalburgi NB, Patil SR, Bhat K. Herpesviruses in chronic and aggressive periodontitis patients in an Indian population. *J Oral Sci* 2009: **51**: 79–86.
14. Blackbourn DJ, Lennette ET, Ambroziak J, Mourich DV, Levy JA. Human herpesvirus 8 detection in nasal secretions and saliva. *J Infect Dis* 1998: **177**: 213–216.
15. Blomlöf L, Hammarström L, Lindskog S. Occurrence and appearance of cementum hypoplasias in localized and generalized juvenile periodontitis. *Acta Odontol Scand* 1986: **44**: 313–320.
16. Bogert M, Berthold P, Brightman V, Craig R, DiRienzo J, Lai CH, Lally E, Oler J, Rams T, Shenker B, Slots J, Taichman N, Tisot R. Longtudinal study of LJP families – two year surveillance. *J Dent Res* 1989: **68** (Special issue): 312.
17. Borrell LN, Beck JD, Heiss G. Socioeconomic disadvantage and periodontal disease: the Dental Atherosclerosis Risk in Communities study. *Am J Public Health* 2006: **96**: 332–339.
18. Botero JE, Parra B, Jaramillo A, Contreras A. Subgingival human cytomegalovirus correlates with increased clinical periodontal parameters and bacterial coinfection in periodontitis. *J Periodontol* 2007: **78**: 2303–2310.
19. Botero JE, Vidal C, Contreras A, Parra B. Comparison of nested polymerase chain reaction (PCR), real-time PCR and viral culture for the detection of cytomegalovirus in subgingival samples. *Oral Microbiol Immunol* 2008: **23**: 239–244.
20. Boudreau S, Hines HC, Hood AF. Dermal abscesses with *Staphylococcus aureus*, cytomegalovirus and acid-fast bacilli in a patient with acquired immunodeficiency syndrome (AIDS). *J Cutan Pathol* 1988: **15**: 53–57.
21. Bruggeman CA. Does cytomegalovirus play a role in atherosclerosis? *Herpes* 2000: **7**: 51–54.

22. Burghelea B, Serb H. Nuclear bodies and virus-like particles in gingival tissue of periodontopathic patients. *Arch Roum Pathol Exp Microbiol* 1990: **49**: 89–92.

23. Burghelea B, Serb H. Ultrastructural evidence of a papovatype viral morphogenesis phenomenon in infiltrating cells from juvenile periodontal lesions. A case report. *Arch Roum Pathol Exp Microbiol* 1990: **49**: 253–267.

24. Caliendo AM, Shahbazian MD, Schaper C, Ingersoll J, Abdul-Ali D, Boonyaratanakornkit J, Pang XL, Fox J, Preiksaitis J, Schönbrunner ER. A commutable cytomegalovirus calibrator is required to improve the agreement of viral load values between laboratories. *Clin Chem* 2009: **55**: 1701–1710.

25. Cao L, Bandelac G, Volgina A, Korostoff J, DiRienzo JM. Role of aromatic amino acids in receptor binding activity and subunit assembly of the cytolethal distending toxin of *Aggregatibacter actinomycetemcomitans. Infect Immun* 2008: **76**: 2812–2821.

26. Capretti MG, Lanari M, Lazzarotto T, Gabrielli L, Pignatelli S, Corvaglia L, Tridapalli E, Faldella G. Very low birth weight infants born to cytomegalovirus-seropositive mothers fed with their mother's milk: a prospective study. *J Pediatr* 2009: **154**: 842–848.

27. Carlsson G, Wahlin YB, Johansson A, Olsson A, Eriksson T, Claesson R, Hänström L, Henter JI. Periodontal disease in patients from the original Kostmann family with severe congenital neutropenia. *J Periodontol* 2006: **77**: 744–751.

28. Caskey MF, Morgan DJ, Porto AF, Giozza SP, Muniz AL, Orge GO, Travassos MJ, Barrón Y, Carvalho EM, Glesby MJ. Clinical manifestations associated with HTLV type I infection: a cross-sectional study. *AIDS Res Hum Retroviruses* 2007: **23**: 365–371.

29. Chalabi M, Moghim S, Mogharehabed A, Najafi F, Rezaie F. EBV and CMV in chronic periodontitis: a prevalence study. *Arch Virol* 2008: **153**: 1917–1919.

30. Chang CM, Yu KJ, Mbulaiteye SM, Hildesheim A, Bhatia K. The extent of genetic diversity of Epstein-Barr virus and its geographic and disease patterns: a need for reappraisal. *Virus Res* 2009: **143**: 209–221.

31. Chebbi F, Poveda JD, Suzuki T, Tai H, Yoshie H, el Tenn R, de Saint-Martin J, Guetard D, Hara K, Dupont B, de The G. Search for infectious HIV in gingival crevicular fluid and saliva of advanced AIDS patients with severe periodontitis. *AIDS* 1997: **11**: 927–928.

32. Chen LL, Sun WL, Yan J, Yu ZS. [Correlation between infection of different glycoprotein B genotypes of human cytomegalovirus and human chronic periodontitis]. *Zhonghua Kou Qiang Yi Xue Za Zhi* 2006: **41**: 212–215. (Chinese).

33. Cheng J, Ke Q, Jin Z, Wang H, Kocher O, Morgan JP, Zhang J, Crumpacker CS. Cytomegalovirus infection causes an increase of arterial blood pressure. *PLoS Pathog* 2009; **5**: e1000427.

34. Christersson LA, Albini B, Zambon JJ, Wikesjö UM, Genco RJ. Tissue localization of *Actinobacillus actinomycetemcomitans* in human periodontitis. I. Light, immunofluorescence and electron microscopic studies. *J Periodontol* 1987: **58**: 529–539.

35. Cobb CM, Ferguson BL, Keselyak NT, Holt LA, MacNeill SR, Rapley JW. A TEM/SEM study of the microbial plaque overlying the necrotic gingival papillae of HIV-seropositive, necrotizing ulcerative periodontitis. *J Periodontal Res* 2003: **38**: 147–155.

36. Coll O, Benoist G, Ville Y, Weisman LE, Botet F, Anceschi TW, Greenough A, Gibbs R, Carbonell-Estrany X. Guidelines on CMV congenital infection. *J Perinat Med* 2009: **37**: 433–445.

37. Combs DR, Reilly EA, Dawson DR III, Avdiushko SA, Danaher RJ, Miller CS. Detection of human cytomegalovirus in dental plaque from individual periodontal sites by real-time polymerase chain reaction. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 2008: **106**: 840–844.

38. Conde-Agudelo A, Villar J, Lindheimer M. Maternal infection and risk of preeclampsia: systematic review and metaanalysis. *Am J Obstet Gynecol* 2008; **198**: 7–22. Comment in: *Evid Based Dent* 2008;**9**:46–47.

39. Contreras A, Falkler WA Jr, Enwonwu CO, Idigbe EO, Savage KO, Afolabi MB, Onwujekwe D, Rams TE, Slots J. Human Herpesviridae in acute necrotizing ulcerative gingivitis in children in Nigeria. *Oral Microbiol Immunol* 1997: **12**: 259–265.

40. Contreras A, Mardirossian A, Slots J. Herpesviruses in HIV-periodontitis. *J Clin Periodontol* 2001: **28**: 96–102.

41. Contreras A, Nowzari H, Slots J. Herpesviruses in periodontal pocket and gingival tissue specimens. *Oral Microbiol Immunol* 2000: **15**: 15–18.

42. Contreras A, Slots J. Herpesviruses in human periodontal disease. *J Periodontal Res* 2000: **35**: 3–16.

43. Contreras A, Slots J. Typing of herpes simplex virus from human periodontium. *Oral Microbiol Immunol* 2001: **16**: 63–64.

44. Contreras A, Umeda M, Chen C, Bakker I, Morrison JL, Slots J. Relationship between herpesviruses and adult periodontitis and periodontopathic bacteria. *J Periodontol* 1999: **70**: 478–484.

45. Contreras A, Zadeh HH, Nowzari H, Slots J. Herpesvirus infection of inflammatory cells in human periodontitis. *Oral Microbiol Immunol* 1999: **14**: 206–212.

46. Cool CD, Rai PR, Yeager ME, Hernandez-Saavedra D, Serls AE, Bull TM, Geraci MW, Brown KK, Routes JM, Tuder RM, Voelkel NF. Expression of human herpesvirus 8 in primary pulmonary hypertension. *N Engl J Med* 2003: **349**: 1113–1122.

47. Corrado E, Novo S. Role of inflammation and infection in vascular disease. *Acta Chir Belg* 2005: **105**: 567–579.

48. Cutilli T, Cargini P, Placidi D, Corbacelli A. Necrotizing fasciitis of the maxillofacial region caused by dental infection. A case report and review. *Minerva Stomatol* 2007: **56**: 469–476.

49. Dawson DR 3rd, Wang C, Danaher RJ, Lin Y, Kryscio RJ, Jacob RJ, Miller CS. Real-time polymerase chain reaction to determine the prevalence and copy number of epstein-barr virus and cytomegalovirus DNA in subgingival plaque at individual healthy and periodontal disease sites. *J Periodontol* 2009: **80**: 1133–1140.

50. Dawson DR 3rd, Wang C, Danaher RJ, Lin Y, Kryscio RJ, Jacob RJ, Miller CS. Salivary levels of Epstein–Barr virus DNA correlate with subgingival levels, not severity of periodontitis. *Oral Dis* 2009: **15**: 554–559.

51. DeWitt GV, Cobb CM, Killoy WJ. The acute periodontal abscess: microbial penetration of the soft tissue wall. *Int J Periodontics Restorative Dent* 1985: **5**: 38–51.

52. Ding F, Feng XH, Meng HX, Zhao YB, Zhang L, Lu RF, Chen ZB. [Relationship between herpesviruses and periodontal pathogenic bacteria in subgingival plaque]. *Beijing Da Xue Xue Bao* 2008: **40**: 318–322. (Chinese).

53. do Canto CL, Granato CF, Garcez E, Villas Boas LS, Fink MC, Estevam MP, Pannuti CS. Cytomegalovirus infection in children with Down syndrome in a day-care center in Brazil. *Rev Inst Med Trop Sao Paulo* 2000: **42**: 179–183.

54. Dodd CL, Winkler JR, Heinic GS, Daniels TE, Yee K, Greenspan D. Cytomegalovirus infection presenting as acute periodontal infection in a patient infected with the human immunodeficiency virus. *J Clin Periodontol* 1993: **20**: 282–285.

55. Dowd JB, Aiello AE, Alley DE. Socioeconomic disparities in the seroprevalence of cytomegalovirus infection in the US population: NHANES III. *Epidemiol Infect* 2008: **16**: 1–8.

56. Dowd JB, Zajacova A, Aiello A. Early origins of health disparities: burden of infection, health, and socioeconomic status in U.S. children. *Soc Sci Med* 2009: **68**: 699–707.

57. Enwonwu CO, Falkler WA Jr, Idigbe EO, Afolabi BM, Ibrahim M, Onwujekwe D, Savage O, Meeks VI. Pathogenesis of cancrum oris (noma): confounding interactions of malnutrition with infection. *Am J Trop Med Hyg* 1999: **60**: 223–232.

58. Falkler WA Jr, Enwonwu CO, Idigbe EO. Microbiological understandings and mysteries of noma (cancrum oris). *Oral Dis* 1999: **5**: 150–155.

59. Farghaly AG, Mansour GA, Mahdy NH, Yousri A. Hepatitis B and C virus infections among patients with gingivitis and adult periodontitis: seroprevalence and public health importance. *J Egypt Public Health Assoc* 1998: **73**: 707–735.

60. Farrell S, Ide M, Wilson RF. The relationship between maternal periodontitis, adverse pregnancy outcome and miscarriage in never smokers. *J Clin Periodontol* 2006: **33**: 115–120.

61. Faveri M, Figueiredo LC, Duarte PM, Mestnik MJ, Mayer MPA, Feres M. Microbiological profile of untreated subjects with localized aggressive periodontitis. *J Clin Periodontol* 2009: **36**: 739–749.

62. Ferretti C. Submental space infection presenting in a patient with Fanconi's anaemia. *S Afr J Surg* 1998: **36**: 69–70.

63. Fine DH, Kaplan JB, Kachlany SC, Schreiner HC. How we got attached to *Actinobacillus actinomycetemcomitans*: a model for infectious diseases. *Periodontol 2000* 2006: **42**: 114–157.

64. Fine DH, Markowitz K, Furgang D, Fairlie K, Ferrandiz J, Nasri C, McKiernan M, Gunsolley J. *Aggregatibacter actinomycetemcomitans* and its relationship to initiation of localized aggressive periodontitis: longitudinal cohort study of initially healthy adolescents. *J Clin Microbiol* 2007: **45**: 3859–3869.

65. Fons MP, Flaitz CM, Moore B, Prabhakar BS, Nichols CM, Albrecht T. Multiple herpesviruses in saliva of HIV-infected individuals. *J Am Dent Assoc* 1994: **125**: 713–719.

66. Garant PR, Baer PN, Kilham L. Electron microscopic localization of virions in developing teeth of young hamsters infected with minute virus of mice. *J Dent Res* 1980: **59**: 80–86.

67. Gemmell E, Yamazaki K, Seymour GJ. The role of T cells in periodontal disease: homeostasis and autoimmunity. *Periodontol 2000* 2007: **43**: 14–40.

68. Gomes MF, Teixeira RT, Plens G, Silva MM, Pontes EM, da Rocha JC. Naso-orbicular tissue necrosis by *Streptococcus parasanguis* in a patient with Fanconi anemia: clinical and laboratory aspects. *Quintessence Int* 2004: **35**: 572–576.

69. Grande SR, Imbronito AV, Okuda OS, Lotufo RF, Magalhães MH, Nunes FD. Herpes viruses in periodontal compromised sites: comparison between HIV-positive and -negative patients. *J Clin Periodontol* 2008: **35**: 838–845.

70. Gredmark-Russ S, Dzabic M, Rahbar A, Wanhainen A, Björck M, Larsson E, Michel JB, Söderberg-Nauclér C. Active cytomegalovirus infection in aortic smooth muscle cells from patients with abdominal aortic aneurysm. *J Mol Med* 2009: **87**: 347–356.

71. Grenier G, Gagnon G, Grenier D. Detection of herpetic viruses in gingival crevicular fluid of patients suffering from periodontal diseases: prevalence and effect of treatment. *Oral Microbiol Immunol* 2009: **24**: 506–509.

72. Hakki SS, Aprikyan AA, Yildirim S, Aydinbelge M, Gokalp A, Ucar C, Guran S, Koseoglu V, Ataoglu T, Somerman MJ. Periodontal status in two siblings with severe congenital neutropenia: diagnosis and mutational analysis of the cases. *J Periodontol* 2005: **76**: 837–844.

73. Hai R, Chu A, Li H, Umamoto S, Rider P, Liu F. Infection of human cytomegalovirus in cultured human gingival tissue. *Virol J* 2006: **3**: 84.

74. Hanookai D, Nowzari H, Contreras A, Morrison JL, Slots J. Herpesviruses and periodontopathic bacteria in Trisomy 21 periodontitis. *J Periodontol* 2000: **71**: 376–384.

75. Hart TC, Atkinson JC. Mendelian forms of periodontitis. *Periodontol 2000* 2007: **45**: 95–112.

76. Hermann RM, Füzesi L, Pradier O, Christiansen H, Schmidberger H. Presence of human papillomavirus-18 and Epstein-Barr virus in a squamous cell carcinoma of the tongue in a 20-year-old patient. Case report and review of the current literature. *Cancer Radiother* 2004: **8**: 262–265.

77. Herrera D, Roldan S, Sanz M. The periodontal abscess: a review. *J Clin Periodontol* 2000: **27**: 377–386.

78. Hochman N, Zakay-Rones Z, Shohat H, Ever-Hadani P, Ehrlich J, Schlesinger M, Morag A. Antibodies to cytomegalo and Epstein-Barr viruses in human saliva and gingival fluid. *New Microbiol* 1998: **21**: 131–139.

79. Holmstrup P, Westergaard J. HIV infection and periodontal diseases. *Periodontol 2000* 1998: **18**: 37–46.

80. Hormia M, Willberg J, Ruokonen H, Syrjänen S. Marginal periodontium as a potential reservoir of human papillomavirus in oral mucosa. *J Periodontol* 2005: **76**: 358–363.

81. Horton AL, Boggess KA, Moss KL, Jared HL, Beck J, Offenbacher S. Periodontal disease early in pregnancy is associated with maternal systemic inflammation among African American women. *J Periodontol* 2008: **79**: 1127–1132.

82. Houri-Haddad Y, Wilensky A, Shapira L. T-cell phenotype as a risk factor for periodontal disease. *Periodontol 2000* 2007: **45**: 67–75.

83. Ibeziako SN, Nwolisa CE, Nwaiwu O. Cancrum oris and acute necrotising gingivitis complicating HIV infection in children. *Ann Trop Paediatr* 2003: **23**: 225–226.

84. Imbronito AV, Grande SR, Freitas NM, Okuda O, Lotufo RF, Nunes FD. Detection of Epstein-Barr virus and human

cytomegalovirus in blood and oral samples: comparison of three sampling methods. *J Oral Sci* 2008: **50**: 25–31.

85. Imbronito AV, Okuda OS, Maria de Freitas N, Moreira Lotufo RF, Nunes FD. Detection of herpesviruses and periodontal pathogens in subgingival plaque of patients with chronic periodontitis, generalized aggressive periodontitis, or gingivitis. *J Periodontol* 2008: **79**: 2313–2321.

86. Isegawa Y, Katahira J, Yamanishi K, Sugimoto N. Reactivation of latent human immunodeficiency virus 1 by human herpesvirus 6 infection. *Acta Virol* 2007: **51**: 13–20.

87. Jaramillo A, Arce RM, Herrera D, Betancourth M, Botero JE, Contreras A. Clinical and microbiological characterization of periodontal abscesses. *J Clin Periodontol* 2005: **32**: 1213–1218.

88. Jaskoll T, Abichaker G, Jangaard N, Bringas P Jr, Melnick M. Cytomegalovirus inhibition of embryonic mouse tooth development: a model of the human amelogenesis imperfecta phenocopy. *Arch Oral Biol* 2008: **53**: 405–415.

89. Jimenez LM, Duque FL, Baer PN, Jimenez SB. Necrotizing ulcerative periodontal diseases in children and young adults in Medellin, Colombia, 1965–2000. *J Int Acad Periodontol* 2005: **7**: 55–63.

90. Kamma JJ, Contreras A, Slots J. Herpes viruses and periodontopathic bacteria in early-onset periodontitis. *J Clin Periodontol* 2001: **28**: 879–885.

91. Keijser BJ, Zaura E, Huse SM, van der Vossen JM, Schuren FH, Montijn RC, ten Cate JM, Crielaard W. Pyrosequencing analysis of the oral microflora of healthy adults. *J Dent Res* 2008: **87**: 1016–1020.

92. Kenneson A, Cannon MJ. Review and meta-analysis of the epidemiology of congenital cytomegalovirus (CMV) infection. *Rev Med Virol* 2007: **17**: 253–276.

93. Kilian M, Frandsen EV, Haubek D, Poulsen K. The etiology of periodontal disease revisited by population genetic analysis. *Periodontol 2000* 2006: **42**: 158–179.

94. Klemenc P, Skaleric U, Artnik B, Nograsek P, Marin J. Prevalence of some herpesviruses in gingival crevicular fluid. *J Clin Virol* 2005: **34**: 147–152.

95. Konstantinidis A, Sakellari D, Papa A, Antoniadis A. Real-time polymerase chain reaction quantification of Epstein–Barr virus in chronic periodontitis patients. *J Periodontal Res* 2005: **40**: 294–298.

96. Kshirsagar AV, Craig RG, Moss KL, Beck JD, Offenbacher S, Kotanko P, Klemmer PJ, Yoshino M, Levin NW, Yip JK, Almas K, Lupovici EM, Usvyat LA, Falk RJ. Periodontal disease adversely affects the survival of patients with end-stage renal disease. *Kidney Int* 2009: **75**: 746–751.

97. Kubar A, Saygun I, Özdemir A, Yapar M, Slots J. Real-time polymerase chain reaction quantification of human cytomegalovirus and Epstein–Barr virus in periodontal pockets and the adjacent gingiva of periodontitis lesions. *J Periodontal Res* 2005: **40**: 97–104.

98. Kubar A, Saygun I, Yapar M, Özdemir A, Slots J. Real-time PCR quantification of cytomegalovirus in aggressive periodontitis lesions using TaqMan technology. *J Periodontal Res* 2004: **39**: 81–86.

99. Kuwabara S. Guillain–Barré syndrome: epidemiology, pathophysiology and management. *Drugs* 2004: **64**: 597–610.

100. Lang NP, Joss A, Tonetti MS. Monitoring disease during supportive periodontal treatment by bleeding on probing. *Periodontol 2000* 1996: **12**: 44–48.

101. Li Y, Zhang JC, Zhang YH. [The association between infection of Epstein–Barr virus and chronic periodontitis]. *Zhonghua Kou Qiang Yi Xue Za Zhi* 2004: **39**: 146–148. (Chinese).

102. Ling LJ, Ho CC, Wu CY, Chen YT, Hung SL. Association between human herpesviruses and the severity of periodontitis. *J Periodontol* 2004: **75**: 1479–1485.

103. Listgarten MA, Slots J, Rosenberg J, Nitkin L, Sullivan P, Oler J. Clinical and microbiological characteristics of treated periodontitis patients on maintenance care. *J Periodontol* 1989: **60**: 452–459.

104. Lundgren T, Parhar RS, Renvert S, Tatakis DN. Impaired cytotoxicity in Papillon–Lefèvre syndrome. *J Dent Res* 2005: **84**: 414–417.

105. Madinier I, Doglio A, Cagnon L, Lefebvre JC, Monteil RA. Epstein–Barr virus DNA detection in gingival tissues of patients undergoing surgical extractions. *Br J Oral Maxillofac Surg* 1992: **30**: 237–243.

106. Madinier I, Doglio A, Cagnon L, Lefèbvre JC, Monteil RA. Southern blot detection of human papillomaviruses (HPVs) DNA sequences in gingival tissues. *J Periodontol* 1992: **63**: 667–673.

107. Mardirossian A, Contreras A, Navazesh M, Nowzari H, Slots J. Herpesviruses 6, 7 and 8 in HIV- and non-HIV-associated periodontitis. *J Periodontal Res* 2000: **35**: 278–284.

108. Matičič M, Poljak M, Kramar B, Seme K, Brinovec V, Meglic-Volkar J, Zakotnik B, Skaleric U. Detection of hepatitis C virus RNA from gingival crevicular fluid and its relation to virus presence in saliva. *J Periodontol* 2001: **72**: 11–16.

109. Matičič M, Poljak M, Kramar B, Tomazic J, Vidmar L, Zakotnik B, Skaleric U. Proviral HIV-1 DNA in gingival crevicular fluid of HIV-1-infected patients in various stages of HIV disease. *J Dent Res* 2000: **79**: 1496–1501.

110. McMillan BC, Golubjatnikov R, Hanson RP, Sinha SK. A study of cytomegalovirus, Epstein–Barr virus and herpesvirus hominis (types 1 and 2) antibody in institutionalized and non-institutionalized children. *Health Lab Sci* 1977: **14**: 261–268.

111. Mendieta C, Miranda J, Brunet LI, Gargallo J, Berini L. Alveolar bone necrosis and tooth exfoliation following herpes zoster infection: a review of the literature and case report. *J Periodontol* 2005: **76**: 148–153.

112. Meron MK, Amital H, Shepshelovich D, Barzilai O, Ram M, Anaya JM, Gerli R, Nicola B, Shoenfeld Y. Infectious aspects and the etiopathogenesis of rheumatoid arthritis. *Clin Rev Allergy Immunol* (published online. doi: 10.1007/s12016-009-8158-6).

113. Michalowicz BS, Durand R. Maternal periodontal disease and spontaneous preterm birth. *Periodontol 2000* 2007: **44**: 103–112.

114. Michalowicz BS, Hodges JS, DiAngelis AJ, Lupo VR, Novak MJ, Ferguson JE, Buchanan W, Bofill J, Papapanou PN, Mitchell DA, Matseoane S, Tschida PA, OPT Study. Treatment of periodontal disease and the risk of preterm birth. *N Engl J Med* 2006: **355**: 1885–1894.

115. Michalowicz BS, Ronderos M, Camara-Silva R, Contreras A, Slots J. Human herpesviruses and *Porphyromonas gingivalis* are associated with early-onset periodontitis. *J Periodontol* 2000: **71**: 981–988.

116. Miller CS, Berger JR, Mootoor Y, Avdiushko SA, Zhu H, Kryscio RJ. High prevalence of multiple human herpes-

viruses in saliva from human immunodeficiency virus-infected persons in the era of highly active antiretroviral therapy. *J Clin Microbiol* 2006: **44**: 2409–2415.

117. Miller GE, Freedland KE, Duntley S, Carney RM. Relation of depressive symptoms to C-reactive protein and pathogen burden (cytomegalovirus, herpes simplex virus, Epstein–Barr virus) in patients with earlier acute coronary syndromes. *Am J Cardiol* 2005: **95**: 317–321.

118. Mocarski ES Jr. Immune escape and exploitation strategies of cytomegaloviruses: impact on and imitation of the major histocompatibility system. *Cell Microbiol* 2004: **6**: 707–717.

119. Mocarski ES Jr, Shenk T, Pass RF. Cytomegaloviruses. In: Knipe DM, Howley PM, editors. *Fields virology*, 5th edn. Philadelphia: Lippincott, Williams & Wilkins, 2007: 2702–2772.

120. Mogensen TH, Paludan SR. Molecular pathways in virus-induced cytokine production. *Microbiol Mol Biol Rev* 2001: **65**: 131–150.

121. Moghim SH, Chalabi M, Abed AM, Rezaei F, Tamizifar H. Prevalence of Epstein–Barr virus type 1 in patients with chronic periodontitis by nested-PCR. *Pak J Biol Sci* 2007: **10**: 4547–4550.

122. Mombelli A, Meier C. On the symmetry of periodontal disease. *J Clin Periodontol* 2001: **28**: 741–745.

123. Moutsopoulos NM, Madianos PN. Low-grade inflammation in chronic infectious diseases: paradigm of periodontal infections. *Ann N Y Acad Sci* 2006: **1088**: 251–264.

124. Muhlestein JB, Anderson JL. Chronic infection and coronary artery disease. *Cardiol Clin* 2003: **21**: 333–362.

125. Müller HP. [Does chronic periodontitis play a role in the pathogenesis of cardiovascular and cerebrovascular diseases?]. *Gesundheitswesen* 2002: **64**: 89–98. (In German).

126. Murata H, Nih R, Ito M, Ihara T, Komada Y. Quantitative detection of HCMV DNA in saliva from infants and breast milk by real time PCR assay. *Pediatr Int* 2009: **51**: 530–534.

127. Nares S. The genetic relationship to periodontal disease. *Periodontol 2000* 2003: **32**: 36–49.

128. Ndiaye FC, Bourgeois D, Leclercq MH, Berthe O. Noma: public health problem in Senegal and epidemilogical surveillance. *Oral Dis* 1999: **5**: 163–166.

129. Nigro G. Maternal-fetal cytomegalovirus infection: from diagnosis to therapy. *J Matern Fetal Neonatal Med* 2009: **22**: 169–174.

130. Nishiyama SA, Nakano V, Velásquez-Melendez G, Avila-Campos MJ. Occurrence of herpes simplex virus 1 and three periodontal bacteria in patients with chronic periodontitis and necrotic pulp. *Can J Microbiol* 2008: **54**: 326–330.

131. Novak MJ, Novak KF, Hodges JS, Kirakodu S, Govindaswami M, Diangelis A, Buchanan W, Papapanou PN, Michalowicz BS. Periodontal bacterial profiles in pregnant women: response to treatment and associations with birth outcomes in the obstetrics and periodontal therapy (OPT) study. *J Periodontol* 2008: **79**: 1870–1879.

132. Nowzari H, Botero JE, DeGiacomo M, Villacres MC, Rich SK. Microbiology and cytokine levels around healthy dental implants and teeth. *Clin Implant Dent Relat Res* 2008: **10**: 166–173.

133. Nowzari H, Jorgensen MG, Ta TT, Contreras A, Slots J. Aggressive periodontitis associated with Fanconi's anemia. A case report. *J Periodontol* 2001: **72**: 1601–1606.

134. Page RC, Offenbacher S, Schroeder HE, Seymour GJ, Kornman KS. Advances in the pathogenesis of periodontitis: summary of developments, clinical implications and future directions. *Periodontol 2000* 1997: **14**: 216–248.

135. Pallasch TJ, Slots J. Oral microorganisms and cardiovascular disease. *J Cal Dent Assoc* 2000: **28**: 204–214.

136. Paraskevas S, Huizinga JD, Loos BG. A systematic review and meta-analyses on C-reactive protein in relation to periodontitis. *J Clin Periodontol* 2008: **35**: 277–290.

137. Parra B, Slots J. Detection of human viruses in periodontal pockets using polymerase chain reaction. *Oral Microbiol Immunol* 1996: **11**: 289–293.

138. Pass RF. Epidemiology and transmission of cytomegalovirus. *J Infect Dis* 1985: **152**: 243–248.

139. Pass RF, Zhang C, Evans A, Simpson T, Andrews W, Huang ML, Corey L, Hill J, Davis E, Flanigan C, Cloud G. Vaccine prevention of maternal cytomegalovirus infection. *N Engl J Med* 2009: **360**: 1191–1199.

140. Paster BJ, Dewhirst FE. Molecular microbial diagnosis. *Periodontol 2000* 2009: **51**: 38–44.

141. Paster BJ, Olsen I, Aas JA, Dewhirst FE. The breadth of bacterial diversity in the human periodontal pocket and other oral sites. *Periodontol 2000* 2006: **42**: 80–87.

142. Paster BJ, Russell MK, Alpagot T, Lee AM, Boches SK, Galvin JL, Dewhirst FE. Bacterial diversity in necrotizing ulcerative periodontitis in HIV-positive subjects. *Ann Periodontol* 2002: **7**: 8–16.

143. Paquette DW, Brodala N, Nichols TC. Cardiovascular disease, inflammation, and periodontal infection. *Periodontol 2000* 2007: **44**: 113–126.

144. Pischon N, Pischon T, Kröger J, Gülmez E, Kleber BM, Bernimoulin JP, Landau H, Brinkmann PG, Schlattmann P, Zernicke J, Buttgereit F, Detert J. Association among rheumatoid arthritis, oral hygiene, and periodontitis. *J Periodontol* 2008: **79**: 979–986.

145. Powers C, DeFilippis V, Malouli D, Früh K. Cytomegalovirus immune evasion. *Curr Top Microbiol Immunol* 2008: **325**: 333–359.

146. Pradeep AR, Hadge P, Arjun Raju P, Shetty SR, Shareef K, Guruprasad CN. Periodontitis as a risk factor for cerebrovascular accident: a case-control study in the Indian population. *J Periodontal Res* 2010: **45**: 223–228.

147. Puchhammer-Stöckl E, Görzer I. Cytomegalovirus and Epstein–Barr virus subtypes – the search for clinical significance. *J Clin Virol* 2006: **36**: 239–248.

148. Rams TE, Listgarten MA, Slots J. Utility of radiographic crestal lamina dura for predicting periodontitis disease-activity. *J Clin Periodontol* 1994: **21**: 571–576.

149. Reddy MS. Reaching a better understanding of non-oral disease and the implication of periodontal infections. *Periodontol 2000* 2007: **44**: 9–14.

150. Reeves M, Sinclair J. Aspects of human cytomegalovirus latency and reactivation. *Curr Top Microbiol Immunol* 2008: **325**: 297–313.

151. Rickinson AB, Kieff E. Epstein–Barr virus. In: Knipe DM, Howley PM, editors. *Fields virology*, 5th edn. Philadelphia: Lippincott, Williams & Wilkins, 2007: 2656–2700.

152. Rist M, Smith C, Bell MJ, Burrows SR, Khanna R. Cross-recognition of HLA DR4 alloantigen by virus-specific CD8+ T cells: a new paradigm for self/non-self recognition. *Blood* 2009: **114**: 2244–2253.

153. Ritter JT, Tang-Feldman YJ, Lochhead GR, Estrada M, Lochhead S, Yu C, Ashton-Sager A, Tuteja D, Leutenegger C, Pomeroy C. In vivo characterization of cytokine profiles and viral load during murine cytomegalovirus-induced acute myocarditis. *Cardiovasc Pathol* 2010: **19**: 83–93.

154. Ross DS, Dollard SC, Victor M, Sumartojo E, Cannon MJ. The epidemiology and prevention of congenital cytomegalovirus infection and disease: activities of the Centers for Disease Control and Prevention Workgroup. *J Womens Health (Larchmt)* 2006: **15**: 224–229.

155. Ross DS, Victor M, Sumartojo E, Cannon MJ. Women's knowledge of congenital cytomegalovirus: results from the 2005 HealthStyles survey. *J Womens Health (Larchmt)* 2008: **17**: 849–858.

156. Rotola A, Cassai E, Farina R, Caselli E, Gentili V, Lazzarotto T, Trombelli L. Human herpesvirus 7, Epstein–Barr virus and human cytomegalovirus in periodontal tissues of periodontally diseased and healthy subjects. *J Clin Periodontol* 2008: **35**: 831–837.

157. Rotundo R, Maggi F, Nieri M, Muzzi L, Bendinelli M, Prato GP. TT virus infection of periodontal tissues: a controlled clinical and laboratory pilot study. *J Periodontol* 2004: **75**: 1216–1220.

158. Saleh A, Stephen LX. Oral manifestations of Fanconi's anaemia: a case report. *SADJ* 2008: **63**: 28–31.

159. Saygun I, Kubar A, Özdemir A, Slots J. Periodontitis lesions are a source of salivary cytomegalovirus and Epstein–Barr virus. *J Periodontal Res* 2005: **40**: 187–191.

160. Saygun I, Kubar A, Özdemir A, Yapar M, Slots J. Herpesviral–bacterial interrelationships in aggressive periodontitis. *J Periodontal Res* 2004: **39**: 207–212.

161. Saygun I, Kubar A, Şahin S, Sener K, Slots J. Quantitative analysis of association between herpesviruses and bacterial pathogens in periodontitis. *J Periodontal Res* 2008: **43**: 352–359.

162. Saygun I, Sahin S, Muşabak U, Enhoş S, Kubar A, Günhan O, Slots J. Human cytomegalovirus in peripheral giant cell granuloma. *Oral Microbiol Immunol* 2009: **24**: 408–410.

163. Saygun I, Yapar M, Özdemir A, Kubar A, Slots J. Human cytomegalovirus and Epstein–Barr virus type 1 in periodontal abscesses. *Oral Microbiol Immunol* 2004: **19**: 83–87.

164. Sculley TB, Apolloni A, Hurren L, Moss DJ, Cooper DA. Coinfection with A- and B-type Epstein–Barr virus in human immunodeficiency virus-positive subjects. *J Infect Dis* 1990: **162**: 643–648.

165. Sedghizadeh PP, Kumar KSS, Gorur A, Schaudinn C, Shuler CF, Costerton JW. Identification of microbial biofilms in osteonecrosis of the jaws secondary to bisphosphonate therapy. *J Oral Maxillofac Surg* 2008: **66**: 767–775.

166. Seidman DS, Samueloff A, Mor-Yosef S, Schenker JG. The effect of maternal age and socioeconomical background on neonatal outcome. *Int J Gynaecol Obstet* 1990: **33**: 7–12.

167. Seymour GJ, Ford PJ, Cullinan MP, Leishman S, Yamazaki K. Relationship between periodontal infections and systemic disease. *Clin Microbiol Infect* 2007: **13** (Suppl. 4): 3–10.

168. Sigusch BW, Wutzler A, Nietzsch T, Glockmann E. Evidence for a specific crevicular lymphocyte profile in aggressive periodontitis. *J Periodontal Res* 2006: **41**: 391–396.

169. Simanek AM, Dowd JB, Aiello AE. Persistent pathogens linking socioeconomic position and cardiovascular disease in the US. *Int J Epidemiol* 2009: **38**: 775–787.

170. Skrepcinski FB, Tetrev S, Rams TE, Sutton B, Contreras A, Slots J. Periodontal disease in Hopi native American teenagers. *J Dent Res* 1997: **76** (special issue): 439 (Abstr 3406).

171. Slots J. Subgingival microflora and periodontal disease. *J Clin Periodontol* 1979: **6**: 351–382.

172. Slots J. Casual or causal relationship between periodontal infection and non-oral disease? *J Dent Res* 1998: **77**: 1764–1765.

173. Slots J. *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis* in periodontal disease: introduction. *Periodontol 2000* 1999: **20**: 7–13.

174. Slots J. Selection of antimicrobial agents in periodontal therapy. *J Periodontal Res* 2002: **37**: 389–398.

175. Slots J. Systemic antibiotics in periodontics. *J Periodontol* 2004: **75**: 1553–1565.

176. Slots J. Herpesviruses in periodontal diseases. *Periodontol 2000* 2005: **38**: 33–62.

177. Slots J. Herpesviral–bacterial synergy in the pathogenesis of human periodontitis. *Curr Opin Infect Dis* 2007: **20**: 278–283.

178. Slots J. Oral viral infections of adults. *Periodontol 2000* 2009: **49**: 60–86.

179. Slots J. Herpesviral–bacterial interactions in periodontal diseases. *Periodontol 2000* 2010: **52**: 117–140.

180. Slots J, Kamma JJ, Sugar C. The herpesvirus-*Porphyromonas gingivalis*-periodontitis axis. *J Periodontal Res* 2003: **38**: 318–323.

181. Slots J, Saygun I, Sabeti M, Kubar A. Epstein–Barr virus in oral diseases. *J Periodontal Res* 2006: **41**: 235–244.

182. Slots J, Schonfeld SE. *Actinobacillus actinomycetemcomitans* in localized juvenile periodontitis. In: Hamada S, Holt SC, McGhee RJ, editors. *Periodontal disease: pathogens and host immune responses*. Tokyo: Quintessence Publishing Co, 1991: 53–64.

183. Slots J, Sugar C, Kamma JJ. Cytomegalovirus periodontal presence is associated with subgingival *Dialister pneumosintes* and alveolar bone loss. *Oral Microbiol Immunol* 2002: **17**: 369–374.

184. Slots J, Ting M. *Actinobacillus actinomycetemcomitans* and *Porphyromonas gingivalis* in human periodontal disease: occurrence and treatment. *Periodontol 2000* 1999: **20**: 82–121.

185. Socransky SS, Haffajee AD. Periodontal microbial ecology. *Periodontol 2000* 2005: **38**: 135–187.

186. Söder B, Jin LJ, Klinge B, Söder PO. Periodontitis and premature death: a 16-year longitudinal study in a Swedish urban population. *J Periodontal Res* 2007: **42**: 361–366.

187. Söderberg-Nauclér C. Does cytomegalovirus play a causative role in the development of various inflammatory diseases and cancer? *J Intern Med* 2006: **259**: 219–246.

188. Söderberg-Nauclér C, Dzabic M, Rahbar A. Understanding the molecular mechanisms involved in indirect effects of cytomegalovirus. *Trends Transplant* 2008: **2**: 32–42.

189. Soto-Ramirez LE, Garcia-Vallejo F, Renjifo B, Vergara A, Borrero I, Marlink R, Essex M. Human T-lymphotropic virus type I (HTLV-I)-specific antibodies and cell-free RNA in crevicular fluid-rich saliva from patients with tropical spastic paraparesis / HTLV-I-associated myelopathy. *Viral Immunol* 1995: **8**: 141–150.

190. Stagno S, Pass RF, Thomas JP, Navia JM, Dworsky ME. Defects of tooth structure in congenital cytomegalovirus infections. *Pediatrics* 1982: **69**: 646–648.

191. Staras SA, Flanders WD, Dollard SC, Pass RF, McGowan JE Jr, Cannon MJ. Cytomegalovirus seroprevalence and childhood sources of infection: a population-based study among pre-adolescents in the United States. *J Clin Virol* 2008: **43**: 266–271.

192. Stenfors LE, Bye HM, Raisanen S, Myklebust R. Bacterial penetration into tonsillar surface epithelium during infectious mononucleosis. *J Laryngol Otol* 2000: **114**: 848–852.

193. Stern J, Shai E, Zaks B, Halabi A, Houri-Haddad Y, Shapira L, Palmon A. Reduced expression of gamma interferon in serum and marked lymphoid depletion induced by *Porphyromonas gingivalis* increase murine morbidity and mortality due to cytomegalovirus infection. *Infect Immun* 2004: **72**: 5791–5798.

194. Sun XJ, Meng HX, Shi D, Xu L, Zhang L, Chen ZB, Feng XH, Lu RF, Ren XY. Elevation of C-reactive protein and inter-leukin-6 in plasma of patients with aggressive periodontitis. *J Periodontal Res* 2009: **44**: 311–316.

195. Sunde PT, Olsen I, Enersen M, Beiske K, Grinde B. Human cytomegalovirus and Epstein–Barr virus in apical and marginal periodontitis: a role in pathology? *J Med Virol* 2008: **80**: 1007–1011.

196. Sunde PT, Olsen I, Enersen M, Grinde B. Patient with se-vere periodontitis and subgingival Epstein–Barr virus treated with antiviral therapy. *J Clin Virol* 2008: **42**: 176–178.

197. Szarka K, Tar I, Fehér E, Gáll T, Kis A, Tóth ED, Boda R, Márton I, Gergely L. Progressive increase of human papil-lomavirus carriage rates in potentially malignant and malignant oral disorders with increasing malignant po-tential. *Oral Microbiol Immunol* 2009: **24**: 314–318.

198. Tabanella G, Nowzari H. Cytomegalovirus-associated periodontitis and Guillain–Barré syndrome. *J Periodontol* 2005: **76**: 2306–2311.

199. Takoudes TG, Haddad J Jr. Retropharyngeal abscess and Epstein–Barr virus infection in children. *Ann Otol Rhinol Laryngol* 1998: **107**: 1072–1075.

200. Tantivanich S, Laohapand P, Thaweeboon S, Desakorn V, Wuthinuntiwong P, Chalermtaranukul S, Pansri P, A-marapal P, Balachandra K, Chantratita W, Dhepakson P. Prevalence of cytomegalovirus, human herpesvirus-6, and Epstein-Barr virus in periodontitis patients and healthy subjects in the Thai population. *Southeast Asian J Trop Med Public Health* 2004: **35**: 635–640.

201. Teughels W, Sliepen I, Quirynen M, Haake SK, Van Eldere J, Fives-Taylor P, Van Ranst M. Human cytomegalovirus enhances *A. actinomycetemcomitans* adherence to cells. *J Dent Res* 2007: **86**: 175–180.

202. Tezal M, Sullivan Nasca M, Stoler DL, Melendy T, Hyland A, Smaldino PJ, Rigual NR, Loree TR. Chronic periodonti-tis-human papillomavirus synergy in base of tongue cancers. *Arch Otolaryngol Head Neck Surg* 2009: **135**: 391–396.

203. Tezal M, Sullivan MA, Reid ME, Marshall JR, Hyland A, Loree T, Lillis C, Hauck L, Wactawski-Wende J, Scannapi-eco FA. Chronic periodontitis and the risk of tongue cancer. *Arch Otolaryngol Head Neck Surg* 2007: **133**: 450–454.

204. Ting M, Contreras A, Slots J. Herpesvirus in localized juvenile periodontitis. *J Periodontal Res* 2000: **35**: 17–25.

205. Toussirot E, Roudier J. Epstein–Barr virus in autoimmune diseases. *Best Pract Res Clin Rheumatol* 2008: **22**: 883–896.

206. Tucker RM, Swanson S, Wenzel RP. Cytomegalovirus and appendiceal perforation in a patient with acquired immunodeficiency syndrome. *South Med J* 1989: **82**: 1056–1057.

207. Umeda M, Contreras A, Chen C, Bakker I, Slots J. The utility of whole saliva to detect the oral presence of periodonto-pathic bacteria. *J Periodontol* 1998: **69**: 828–833.

208. Upadhyay S, Marks SC, Arden RL, Crane LR, Cohn AM. Bacteriology of sinusitis in human immunodeficiency virus-positive patients: implications for management. *Laryngoscope* 1995: **105**: 1058–1060.

209. Van Dyke TE, Vaikuntam J. Neutrophil function and dys-function in periodontal disease. *Curr Opin Periodontol* 1994: 19–27.

210. van Zeeburg HJ, Snijders PJ, Wu T, Gluckman E, Soulier J, Surralles J, Castella M, van der Wal JE, Wennerberg J, Califano J, Velleuer E, Dietrich R, Ebell W, Bloemena E, Joenje H, Leemans CR, Brakenhoff RH. Clinical and molecular characteristics of squamous cell carcinomas from Fanconi anemia patients. *J Natl Cancer Inst* 2008: **100**: 1649–1653.

211. Velazco CH, Coelho C, Salazar F, Contreras A, Slots J, Pacheco JJ. Microbiological features of Papillon–Lefèvre syndrome periodontitis. *J Clin Periodontol* 1999: **26**: 622–627.

212. Vercellotti GM. Overview of infections and cardiovascular diseases. *J Allergy Clin Immunol* 2001: **108** (4 Suppl.): S117–S120.

213. Vilkuna-Rautiainen T, Pussinen PJ, Roivainen M, Petäys T, Jousilahti P, Hovi T, Vartiainen E, Asikainen S. Serum antibody response to periodontal pathogens and herpes simplex virus in relation to classic risk factors of cardio-vascular disease. *Int J Epidemiol* 2006: **35**: 1486–1494.

214. Waldman M, Marshall V, Whitby D, Kopp JB. Viruses and kidney disease: beyond HIV. *Semin Nephrol* 2008: **28**: 595–607.

215. Watanabe SA, de Fátima Correia-Silva J, Horta MC, da Costa JE, Gomez RS. EBV-1 and HCMV in aggressive periodontitis in Brazilian patients. *Braz Oral Res* 2007: **21**: 336–341.

216. Wu YM, Yan J, Chen LL, Sun WL, Gu ZY. Infection fre-quency of Epstein–Barr virus in subgingival samples from patients with different periodontal status and its correla-tion with clinical parameters. *J Zhejiang Univ Sci B* 2006: **7**: 876–883.

217. Wu YM, Yan J, Ojcius DM, Chen LL, Gu ZY, Pan JP. Cor-relation between infections with different genotypes of human cytomegalovirus and Epstein–Barr virus in sub-gingival samples and periodontal status of patients. *J Clin Microbiol* 2007: **45**: 3665–3670. Erratum in: *J Clin Microbiol* 2008: **46**: 836.

218. Yao QY, Tierney RJ, Croom-Carter D, Dukers D, Cooper GM, Ellis CJ, Rowe M, Rickinson AB. Frequency of multiple Epstein–Barr virus infections in T-cell immunocompro-mised individuals. *J Virol* 1996: **70**: 4884–4894.

219. Yildirim S, Yapar M, Kubar A. Detection and quantification of herpesviruses in Kostmann syndrome periodontitis using real-time polymerase chain reaction: a case report. *Oral Microbiol Immunol* 2006: **21**: 73–78.