Does apical periodontitis have systemic consequences? The need for well-planned and carefully conducted clinical studies

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IN BRIEF

- Reports that current investigations of apical periodontitis and its potential systemic consequences show only weak associations between apical periodontitis and certain comorbidities.
- Indicates that previous study designs were such that possible causal relationships could not be exposed.
- Stresses the need for intervention studies and provides suggestions for a good study design.

OPINION

Apical periodontitis, infection of the root canal system, may have systemic consequences. This proposal has been brought forward many times in dentistry literature but the general consensus is that there is no scientific basis for an association between endodontic infections and general health. This opinion paper argues that, in order to obtain such a scientific basis, or to rule out the issue all together, we need carefully designed longitudinal challenge model (that is, intervention) studies in which we follow specific biomarkers of inflammation. These biomarkers can be those that are currently being substantiated in chronic inflammation and low-grade inflammation studies in medicine and nutritional science, where the presence of these inflammatory disorders is linked to systemic outcomes. A list of suggested biomarkers has been included.

Apical periodontitis (AP) is brought about by microbial infection of the root canal system.[1] After invasion by microorganisms, the pulpal tissue becomes necrotic and then the root canal system becomes a haven for microbial biofilms. Subsequently, inflammatory responses occur at the root tip where the microorganisms come in direct contact with the periapical tissues. Bone is resorbed to allow invasion of inflammatory cells, which is when AP becomes visible on a radiograph as a radiolucent area around the root tip. Although severe pain can occur, this process is usually accompanied with mild or no symptoms. The need for treatment is obvious and mandatory in symptomatic AP, but what if AP is incidentally discovered or what if a previous root canal treatment has not resolved the AP? Is one then justified to accept this inflammatory process or is treatment indicated (again)?[2] To answer these questions it is important to establish what happens if AP is left untreated.

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The local consequences of AP have been well studied in root-canal-treated teeth with persistent AP. In these teeth, the prevalence of AP is about 50%.[3,4] The 10-year survival rate on the other hand is high and varies between 81% and 93%.[5–7] Of the lost teeth, only 7–19% were extracted for endodontic reasons.[8,9] We can conclude from these numbers that a chronic AP lesion seldom leads to severe problems such as large bone lesions with tooth mobility and combined periodontal-endodontic lesions, persisting fistulas and possibly cyst formation. This is an important reason why asymptomatic AP is often left untreated. Yet, the potential systemic consequences of AP have been less well studied and whether AP affects general health has occupied the dental profession for decades.

Recently, the Swedish Council on Health Technology Assessment (SBU) has evaluated the methods used by dentists to diagnose, prevent and treat infection and inflammation of the dental pulp.[10] One of the questions that the SBU addressed was: 'Is there a risk that cases of acute and chronic infection originating in the dental pulp may give rise to pathological conditions in other organs?' One of SBU's conclusions was that the scientific basis was insufficient to assess the association between endodontic infections and disease conditions of other organs. When an association was found, then that relationship was weak. Thus, either there is no association between AP and systemic health or previously performed studies have not been designed well enough to demonstrate any relationship. In other areas of medicine, co-morbidities have been associated with inflammation and therefore it is more than likely that a periapical inflammation will have systemic consequences too.[11–13] Endodontic research has mainly focused on associations between AP and disease. This is an important reason why a causal relationship between AP and general health has not been found yet.

In a recent review about AP and its effect on systemic levels of inflammatory markers, the authors did conclude that AP is associated with increased levels of BOIs in humans.[14] In a cross-sectional study for instance, the levels of interleukin (IL)-1, -2 and -6 were raised compared to the healthy controls.[15] In this review however, it was also stated, that the findings were not conclusive regarding the efficacy of endodontic treatment in reducing the serum levels of the different biomarkers. This remarkable finding is likely due to the heterogeneity of methodology of the included studies. There was great variation in study design, the control of potential confounding variables and the choice of BOIs in the different investigations. These variations make comparing or pooling data difficult or impossible. Inspired by recent insights and experimental models that are being developed in nutritional studies,[16] we will present an overview of inflammatory markers that can be used to monitor changes in the systemic inflammatory state. Moreover, a solid study design is proposed.

Apical periodontitis is a local inflammatory response. Inflammation is part of the complex biological response of vascular tissues to harmful stimuli, such as pathogens,

damaged cells, or irritants. It is a dynamic process that lasts from a few minutes to years, depending on the extent of the injury, the type of injury and the vascularity of the tissue. An inflammatory response has several functions: to inactivate the injurious agent, to break down and remove dead tissue and to initiate healing of the tissue. The inflammatory response is a complex response that involves circulatory (haemo-dynamic) changes, changes in vessel wall permeability, response of host defence cells (consisting of white blood cells and other cell types in the tissues) and the release of soluble mediators through several pathways. The resolution of an inflammatory response is an active process with the release of anti-inflammatory and pro-resolving mediators.[17] Three main stages in the inflammatory response can be distinguished. The acute phase has an immediate onset and duration of a few days. Its usual outcome is the resolution of the inflammation with or without abscess formation. In acute inflammation neutrophils, other granulocytes and mononuclear phagocytes (monocytes, macrophages) are the major cells involved. Primary mediators are vaso-active amines and eicosanoids which are released within minutes of stimulation. Chemokines and cytokines, IL-1, tumour necrosis factor (TNF), IL-6, IL-11, IL-8 and other chemokines, granulocyte colony stimulating factor (GCSF), granulocyte chemotactic protein-2 (GCP-2), monocyte chemotactic protein-3 (MCP-3), and granulocyte/monocyte colony stimulating factor (GM-CSF) that play important roles in acute inflammation are generally elevated within a few hours of the inflammatory perturbation. Acute inflammation is a normal physiological response and it is crucial for maintaining homeostasis.

A chronic inflammation occurs when an acute inflammation fails to resolve and can be driven by dysregulated pro-inflammatory processes. Its onset is delayed but it can last for an unlimited time. The major cells involved are mononuclear phagocytes (monocytes, macrophages, osteoclasts), B-lymphocytes and T-lymphocytes, neutrophils and fibroblasts, among others. Chronic inflammatory mediators are predominantly cytokines, chemokines, eicosanoids, growth factors, reactive oxygen species and hydrolytic enzymes. The liver produces acute phase proteins, for instance C-reactive protein, which facilitates complement activation by bacteria and dead and dying host cells. Where inflammation exists, there is uncontrolled or excessive or continued, unabated tissue damage, and metabolic changes occur. A chronic inflammation can exacerbate into episodes of acute inflammation. Moreover, in a chronic inflammatory response, inflammatory mediators (IMs) are continuously expressed to sustain the inflammation. They can spill over into the circulation and exert systemic effects or contribute to existing pathology. IMs can be detected in blood or serum and therefore they may serve as a biomarker of inflammation.

A low-grade inflammation – a currently misused phrase in dentistry – is defined as a serious metabolic disturbance. The primary mediators are essentially the same as in a chronic inflammation. There is however, no overt pathology like vascular or tissue damage. In a low-grade chronic inflammation, increased insulin resistance and intracellular lipid accumulation is seen. There are a number of exogenous and endogenous modifiers that influence the expression of IMs and which can give rise to a low-grade inflammatory state. These modifiers can be age, body weight, physical exercise, recent surgery, diet (sugary and fatty meals give rise to a temporary increase in IMs), emotional stress, gut flora, etc.[16] According to this description of the inflammatory stages, non-painful AP should be considered as a chronic inflammatory condition.

Table 1 Potential biomarkers of inflammation

Type of marker	Example
Acute inflammation	
Vasoactive amines	Histamine, serotonin
Proteins/peptides from acute phase reactants Complement, Kinin system Clotting/Fibrinolytic cascade	CRP, fibrinogen, C3 ,C4, amyloid A, haptoglobin, sialic acid C1, activation enzymes & cleavage products, C3a, C5a, etc Bradykinin Plasminogen activators
Arachidonic acid metabolites	Prostaglandins/leukotrienes PGE_2, D_2, F_2 & I_2, etc/LTA_4, B_4, C_4 and D_4.
Platelet activating factors (PAF)	Acetyl glycerol ether phosphocholine
Integrins; adhesion molecules	α/β integrins; Ig superfamily , selectins, hyaladherins & cadherins
Chemokines	IL-8/CXCL-8 and other chemokines, ie CXCL-1, CXCL-8, GCSF, GCP-2, MCP3, and GM-CSF , MIP-2/CXCL-2, etc
Cytokines	IL-1α/β, tumor necrosis factor (TNF), IL-6, IL-11, IL-12, IL-21, IL-17, interferons- (α, β, γ)
Phagocyte products	Cationic proteins, neutral proteinases, oxygen derived free radicals, nitric oxide (various sources)
Chronic inflammation	
As above	Vasoactive amines, plasma cascades. arachidonic acid metabolites, PAF, adhesion molecules, phagocyte products, etc
Chemokines	IL-8/CXCL-8 and other chemokines, ie GCSF, GCP-2, MCP3, GM-CSF
Cytokines In cell mediated responses:	IL-1α/β, TNF and leukotoxin, IL-6, IL-2, IL-3, IL-4, IL-7, IL-9, IL-10, IL-12, IFNs, IFN-γ inducing factor (IGIF), transforming growth factor- β (TGF- β)
In humoral inflammation:	IL-3, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, MIP-1, TGF- β, IL-1 receptor antagonist (IL-1RA), IL-25/IL-17E
In chronic infectious diseases:	IL-17A and IL-17F. IL-17A/F heterodimer
In osteoimmunology	IL-1α/β, IL-6, TNF, RANK, RANKL, OPG, IL-17A and IL-17F. IL-17A/F heterodimer, etc
Enzymes in tissue Remodelling/destruction	Matrix metalloproteinases: Interstitial collagenase (MMP-1), neutrophil collagenase (MMP-8), collagenase-3 (MMP-13), gelatinase A (MMP-2), gelatinase B (MMP-9) Serine esterases: neutrophil elastase, cathepsin G, complement enzymes
Mediators of inflammation which can potentially be used to monitor changes in acute or chronic inflammatory responses.	

CHOICE OF BIOMARKERS OF INFLAMMATION (BOIS)

There are a number of candidate BOIs from which to choose (Table 1). Chronic inflammatory diseases and low-grade inflammation are characterised by the increase in serum and often in other tissue fluids of acute-phase reactants (CRP, fibrinogen, complement proteins (C3, C4), serum amyloid A,

haptoglobin, sialic acid) and pro-inflammatory and anti-inflammatory cytokines. The cytokines that mediate chronic inflammatory processes can be divided into those that contribute to cellular inflammation: IL-1, IL-2, IL-3, IL-4, IL-7, IL-9, IL-10, IL-12, interferons (IFNs), IFN-γ inducing factor (IGIF), transforming growth factor (TGF), TNF and leukotoxin; those participating in humoral inflammation: IL-3, IL-4, IL-5, IL-6, IL-7, IL-9, IL-10, IL-13, macrophage inhibitory protein-1 (MIP-1), transforming growth factor TGF, IL-1 receptor antagonist (IL-1RA) IL-10; and those also involved in chronic infectious diseases and are crucial in protecting the host from invasion by many types of pathogen such as IL-17A and IL-17F. Dysregulated IL-17A and IL-17F production can result in excessive pro-inflammatory cytokine expression, lead to tissue damage and exacerbate chronic inflammation, and autoimmunity. Their activity is often opposed by IL-25/IL-17E the anti-inflammatory IL-17 family member.[18] Furthermore, a number of mediators are involved in tissue remodelling during health and the disease processes; these consist of enzymes such as the matrix metalloprotenases (MMPs) their inhibitors, chemokines and cytokines (which modulate the functions of cells involved during tissue remodeling). Although osteoblasts are understood to be responsible for bone formation and their activity can be monitored by measuring osteocalcin, they are inextricably linked to bone remodeling and are known to promote osteoclast activity.[19] Since soft tissue and hard tissue is destroyed during the active phase of periapical disease, mediators which modulate cell functions such as the chemokine CXCL12 that attracts osteoclast precursor cells to inflammatory sites and receptor activator of nuclear factor kappa B ligand (RANKL) which promotes osteoclastogenesis and the functions of osteoclasts are strongly implicated in the disease process. Osteoclasts secrete hydroxonium ions directly onto the surface of bone resulting in solubilisation of the bone matrix. These cells also secrete enzymes such as MMP-9 (gelatinase B/Type IV collagenase). Indirect evidence of enzymatic activity is evident in gingival crevicular fluid (GCF) from patients with periapical disease.[20] Evidence for increased bone degradation during the active phase of disease can be determined by measuring the circulating levels of the C-telopeptide pyridinoline cross links of type I collagen (ICTP) as wells as measuring an increased ratio of soluble RANKL (sRANKL) and the RANKL decoy receptor osteoprotegerin (OPG) in the circulation or other tissue fluids such as GCF and saliva.[21]

Low albumin concentrations in serum often accompany the increase in these markers of inflammation.[22–24] Acute-phase reactants are synthesised in the liver, and its production is regulated by cytokines, including IL-6 and TNF.[25] Serum CRP is a very sensitive systemic marker of inflammation, which increases rapidly in response to a variety of stimuli but is present at low concentrations under normal conditions.[26,27]

STUDY DESIGN

The use of a challenge model should give a meaningful indication of the inflammatory state in a population rather than the assessment of markers during steady state.[16] In such a model, the ability of a subject to recover from a challenge would be assessed at multiple sampling points. This way, a pattern can be observed. Comparing the patterns of healthy and AP subjects could give valuable information about the influence of AP on the resilience of a subject. Unfortunately, such models are not available yet. However in a modified challenge model that is, an intervention model, samples can be taken from AP subjects before and after a treatment after which the data should be analysed with dependent statistical tests. This will give information about changes in the background expression of BOIs. Measuring multiple times within one subject should also decrease the risk of confounding factors. Indeed, if endogenous or exogenous modifiers remain stable during the course of the study, they will become part of the background expression. Therefore, each subject is in essence his/her own control and if during the course of the study his/her condition does not change, then the inclusion criteria could be less strict than when data are compared between subjects. Full medical histories and intra-oral examination of study participants will be essential. A medical history should comprise data on gender, age, body mass, physical (in)activity, genetics, pregnancy, smoking, diet, use of medications and other factors, such as emotional stress, pollution, viral infection and sleep behaviour.[16]

CONCLUSION

It is important to assess whether AP has an effect on the expression of particular BOIs because AP may contribute to the development of co-morbidities or existing pathology. The most important markers that indicate the transition of an acute inflammation to a chronic inflammation are the cytokine IL-6 and the acute phase protein CRP. The episodic nature of certain chronic inflammatory diseases is accompanied by changes in the balance in serum and other tissue fluids of pro-inflammatory cytokines such as: TNF, IL-1 IL-17A, IL-17F, anti-inflammatory cytokines TGF-beta, IL-10, IL-25 (IL-17E), cytokines involved in cell mediated immunity against intracellular pathogens (for example, IFN-γ) and mediators of soft tissue remodeling, MMPs, etc, and particularly of notably bone turnover: sRANKL, OPG, ICTP and osteocalcin which also makes these useful molecules to consider as BOIs. In an intervention study design, resolution of AP could be evaluated successfully by the expression of biomarkers of inflammation or the lack thereof. Once this technology has been refined, it will be valuable in the decision making of whether or not to treat a persistent endodontic infection. It may also be a useful (additional) tool to diagnose AP as at present, intra-oral radiographs play an important role in the diagnosis of AP. However, with a sensitivity of only 50% in diagnosing AP, they are not very accurate. Cone beam computed tomography technology (CBCT) is more sensitive, but a much higher dose of ionising radiation is required which tends to argue against a routine use of CBCT.[28]

Apical periodontitis and its possible effect on systemic health is a recurring theme in dentistry. If science is to solve this dilemma then it is necessary that the definition of AP, as well as the tools to evaluate the presence or absence of AP, are precise. Only then, will one be able to generate meaningful and comparable data in the future.

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