

# The Effect of Smoking on Components of Gingival Crevicular Fluid in Patients with Periodontal Disease

Tsiraki Marina, Tsachouridou Ioanna, Mataftsi Maria, Tsalikis Lazaros, Konstantinidis Antonios

Department of Preventive Dentistry, Periodontology and Implant Biology, Aristotle University of Thessaloniki, Greece

## Abstract

**Aim:** Smoking is one of the major environmental risk factors for periodontal diseases. It may be hypothesized that the gingival crevicular fluid profile in periodontal patients who smoke may differ from the profile of non-smokers. The aim of this literature review is to examine if smoking affects the composition of gingival crevicular fluid in patients with chronic periodontitis. **Methods:** Databases were searched from 2000 to 2014 using various combinations of keywords. Titles and abstracts of articles that fulfilled the eligibility criteria were screened by the authors and checked for agreement. Only articles published in English were reviewed.

**Results:** Twelve studies were included. One study reported no differences in IL-1 $\beta$  and IL-1ra between smokers and non-smokers, while another study showed significantly lower concentration of IL-1 $\beta$  in smokers in deep bleeding sites and IL-1ra in all sites. Two studies reported significantly reduced IL-1ra in smokers. One study found significantly less U-PA, IL-6, IL-12 and IL-8 in smokers, while another showed increased myeloperoxidase (GM) in smokers. Two studies reported no significant differences between the groups as for their cytokine and inflammatory mediator release. One study showed different vascular function and another one reported decreased host defense in smokers, while another found the opposite results. **Conclusion:** According to the existing controversial data, an overall uniform conclusion could not be reached about the GCF profile in smokers and non-smokers periodontal patients. It is therefore suggested that further research should be conducted, focusing on the same components of GCF and utilizing the same methods of collection in order to gain more solid knowledge on the similarities or differences of the GCF profile in smokers and non-smokers periodontal patients.

*Key Words: Periodontitis, Periodontal disease, Smoking, GCF*

## Introduction

Smoking is one of the major environmental risk factors for periodontal diseases [1,2]. It increases the risk of developing periodontitis (Odds Ratio 2, 82) [3]. Smokers tend to be more susceptible to advanced and aggressive forms of periodontal diseases [4,5] and are at greater risk of exhibiting more severe bone loss [6,7]. As for the clinical evaluation after treatment, smokers tend to respond less favorably [8-10] and present with persistent periodontitis more frequently [11]. Regarding the periodontal pathogens that colonize the subgingival microflora in smokers, the results are conflicting [12-15]. Thus, researchers suggest that the main effect of cigarette smoking as a risk factor for chronic periodontitis is the alteration of host response, including antibody production, neutrophil/monocyte activities, vascular function and cytokine-inflammatory mediator release [16,17].

Gingival crevicular fluid (GCF) is initially formed as a transudate of interstitial fluid produced by osmotic phenomena in the basement membrane, which then becomes an inflammatory exudate, due to increased permeability of blood vessels after chemical or mechanical stimulation. Thus, GCF has at first a similar concentration as interstitial fluid and while it traverses inflamed tissues it picks up enzymes and other products of cell and tissue degradation, and ends up being a true exudate of serum, as the inflammation proceeds [18,19].

## Aim

The aim of this literature review is to examine if smoking affects the composition of gingival crevicular fluid in patients with chronic periodontitis.

## Materials and Methods

### Focused question

The addressed focused question was “Does smoking affect the composition of GCF in patients with chronic periodontitis?”

### Eligibility criteria

The selection criteria encompassed the following: 1. Original articles; 2. Human studies; 3. Clinical Trials; 4. Meta-Analysis; 5. Randomized Controlled Trials; 6. Reviews; 7. Articles written in English language; 8. Articles published from 2000 to 2014. Experimental studies, letters to the editor, historical reviews, case reports and unpublished articles were excluded.

### Search strategy

MEDLINE/ PubMed (National Library of Medicine), Embase, Cochrane Database and Ovid (via heal-link) were searched for relevant articles, using the following keywords in various combinations: “smoking”, “tobacco”, “gcf”, “gingival crevicular fluid”, “periodontal disease” and “periodontitis”. A hand search was also performed. Titles and abstracts of articles that satisfied the eligibility criteria were screened by the authors and checked for agreement. The full-text of the articles judged by the title and abstract to be relevant were read and independently assessed against the selection protocol. Any disagreement between the authors was resolved via discussion.

The studies that were finally included in the present review were controlled for confounding parameters, including age, smoking, body mass index, medication, systemic health conditions and alcohol consumption. Letters to the editor, historic reviews and unpublished articles were excluded. The initial search yielded 101 articles in Pubmed, 177 articles in Embase, 15 articles in Cochrane Database and 15 articles in

Ovid. After deleting the diplotypes and screening the full text for suitability, twelve studies were finally selected and processed for data extraction (Figure 1).

### Results

#### Outcome

The selected papers are summarized in Table 1. Table 1 also provides a short summary of the patient population, the smoking status, the biological parameters that were investigated and the results of each study.

Boström et al. [20] studied the effect of smoking on the GCF levels of interleukin-1β (IL-1β) and its receptor antagonist IL-1ra in patients with moderate-to-severe periodontitis. Mean level of IL-1β and IL-1ra was not found to be different between smokers and non-smokers. Similarly, Erdemir et al. [21] found no differences in IL-6 and tumor necrosis factor (TNF-a) (3<sup>rd</sup> and 6<sup>th</sup> month of observation). In contrast to the above studies, Rawlinson et al. [22] found statistically significant differences for both IL-1β and IL-1ra in deep bleeding sites between smokers and non-smokers and

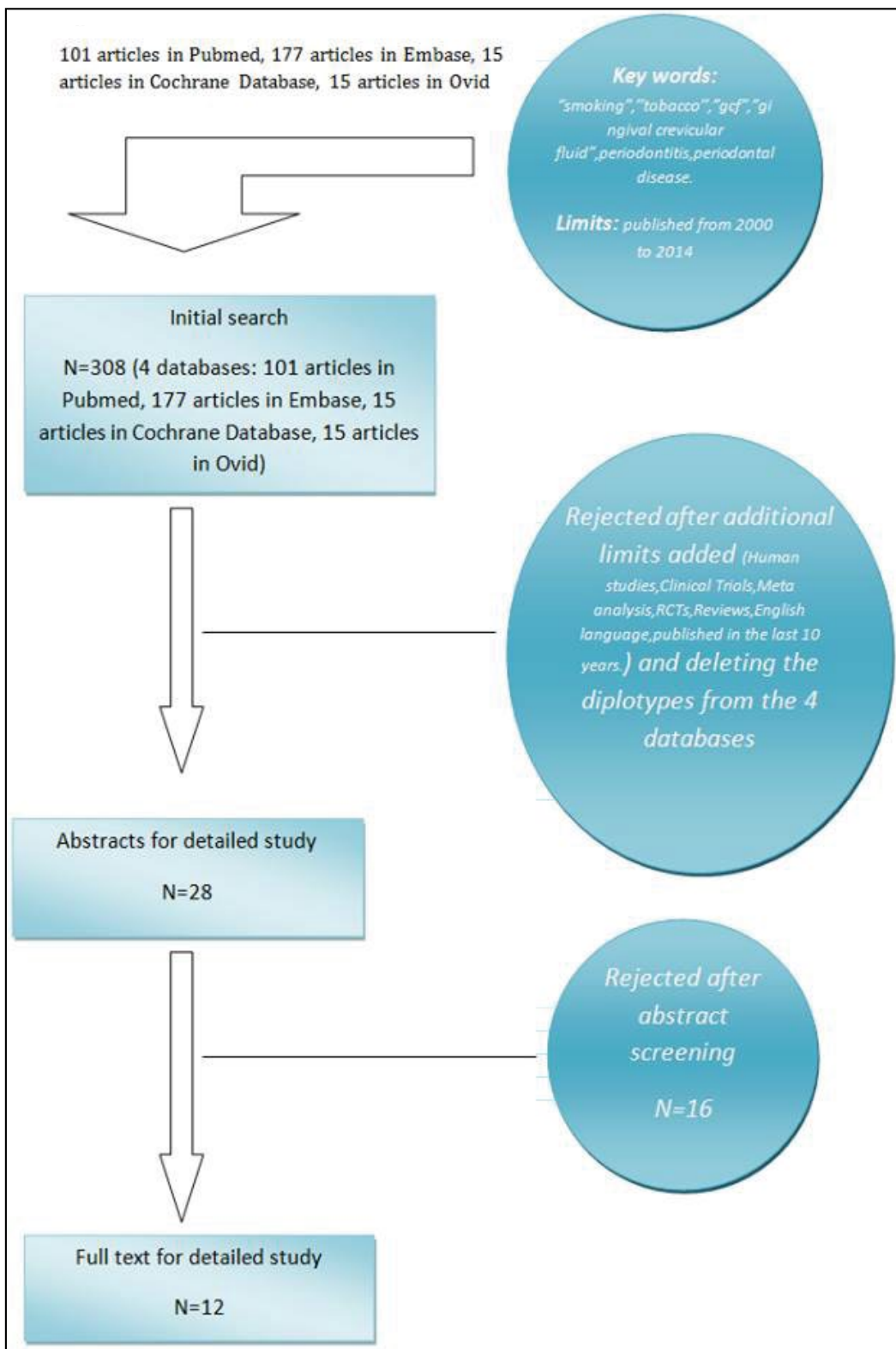


Figure 1. 101 articles in PubMed, 177 articles in Embase, 15 articles in Cochrane Database, 15 articles in Ovid.

Table 1. Summary of selected papers.

Authors (publication date)	Groups	Smoking status	GCF sampling	Biological parameter	Determination of the biological parameter	Results
Boström et al. 2000	40 subjects • 22 smokers • 18 non smokers	Smokers: 20 cigarettes/day>31years Non smokers: never smoked	Aspiration method	IL-1 $\beta$ , IL-1ra	ELISA	No significant differences either in IL-1b or IL-1ra between smokers and non smokers(p>0,05)
Fraser et al. 2001	28 subjects • 14 smokers • 14 non smokers	Smokers : $\geq$ 10 cigarettes/day $\geq$ 3 years Non smokers: no cigarettes in the previous 10 years	Periopaper strip into the gingival crevice for 30 sec	s ICAM-1	ELISA	s-ICAM1:significantly lower in smokers compared with non smokers(p<0,05)
Rawlinson et al. 2003	23 subjects • 13 smokers • 10 non smokers	Smokers :15-79 pack years(mean 32,5 $\pm$ 17.75)	Periopaper strip 1mm into the gingival crevice or pocket entrance for 3 min	IL-1 $\beta$ , IL- 1ra	ELISA	IL-1b: significantly lower concentration in smokers for deep bleeding sites(p<0.05) IL-1ra: significantly lower concentration in smokers for healthy, deep non-bleeding and deep bleeding sites
Erdemir et al. 2004	41 subjects • 22 smokers • 19 non smokers	Smokers : 20 cigarettes/day Non smokers: never smoked	Periopaper strip into the gingival crevice for 30 sec	IL- 6, TNF- $\alpha$	ELISA	No significant differences between smokers and non smokers in the concentration and total amounts of IL-6 and TNF-a both in the 3 <sup>rd</sup> and 6 <sup>th</sup> month after non-surgical periodontal therapy(p>0,05)
Petropoulos et al. 2004	33 subjects • 14 smokers • 19 non smokers	Smokers :not defined Non smokers: never smoked or ex-smokers having stopped at least 2years before	Durapore strip in the pocket for 10 sec	IL- 1 <sup>a</sup>	ELISA	IL-1a: significantly reduced in smokers(p<0,05)
Buduneli et al. 2005	60 subjects • 20 patients with chronic gingivitis (10 smokers,10 non smokers) • 20 patients with chronic periodontitis (10 smokers, 10 non smokers) • 20 periodontally healthy adults (10 smokers, 10 non smokers)	Smokers : $\geq$ 10 cigarettes/day>5years Non smokers: never smoked	Periopaper strip in the orifices of gingival sulcus/pocket for 30 sec	Plasminogen activator system proteins( PA)	ELISA	• No significant differences in t-PA and PAI-1 level and the ratios of PAs to PAIs between smokers and non smokers (p>0,05) • u-PA level: significantly lower in smokers (p<0,05)
Kurtis et al. 2007a	42 subjects • 21 smokers with chronic periodontitis • 21 non smokers with chronic periodontitis	Smokers: $\geq$ 10 cigarettes/day>5yars Non smokers: never smoked	Periopapaer strip into the gingival crevice $\leq$ 1mm for 30 sec	PGE <sub>2</sub> - TBARS	ELISA	• No significant differences in PGE2, TBARS levels between smokers and non smokers at baseline(p>0,05) • No significant differences in PGE2 levels between smokers and non smokers after therapy(p>0,05) • No significant differences in TBARS levels between groups 2 and 4(p>0,05) after therapy • Significantly lower TBARS levels in group 3(smokers) compared with group 1 (non smokers) after therapy(p<0,05)
Kurtis et al. 2007b	58 subjects • 29 smokers with chronic periodontitis • 29 non smokers with chronic periodontitis	Smokers : $\geq$ 10 cigarettes/day>5yars Non smokers: never smoked	Periopaper strip into the gingival crevice $\leq$ 1mm for 30 sec	metalloproteinases 8 (MMP- 8)	ELISA	No significant differences in MMP-8 levels between smokers and non smokers at baseline anafter therapy(p>0,05)
Buduneli et al. 2009	20 subjects • 10 smokers with chronic periodontitis • 10 non smokers with chronic periodontitis	Smokers : $\geq$ 10 cigarettes/day>years Non smokers: never smoked	Periopapaer strip in the gingical sulcus/pocket for 30 sec	IL- 17, receptor activator of nuclear factorkappa B ligand (sRANKL) , osteoprotegerin (OPG)	ELISA	No significant differences in the levels of IL-17, s RANKL, OPG between smokers and non smokers at baseline and 4 weeks after treatment(p>0,05)

Rai et al. 2010	45 subjects <ul style="list-style-type: none"> <li>12 smokers with chronic periodontitis</li> <li>10 non smokers with chronic periodontitis</li> <li>11 smokers with healthy periodontium</li> <li>12 non smokers with healthy periodontium</li> </ul>	Smokers : >18-20 cigarettes/day Non smokers: never smoked	Periopaper strip in gingival pocket for 45 sec	GM	Not reported	GM: significantly increased in smokers (p<0,05)
Tymkiw et al. 2011	52 subjects <ul style="list-style-type: none"> <li>20 smokers with chronic periodontitis</li> <li>20 non smokers with chronic periodontitis</li> <li>12 periodontally healthy controls</li> </ul>	Smokers : ≥20 cigarettes/day Non smokers: not having smoked 100 or more cigarettes in their lifetime	Periopaper strip into the crevice 1-2 mm for 30 sec	IL-2, IL-12(p70), IFN-γ, IL-3, IL-4, IL-5, IL-10, IL-1b, IL-6, GM-CSF, TNF-a-IL-12, IL-8, IP-10, MCP-1, MIP-1a, RANTES, Eotaxin, IL-7, IL-15	Millipore in the Luminex100 IS Instrument	<ul style="list-style-type: none"> <li>Healthy sites in smokers (SH): significantly less IL-6, IL-12 (p40) compared with healthy sites in non smokers (NH)</li> <li>Diseased sites in smokers (SD): significantly less IL-1a, IL-6, IL-12 compared with diseased sites in non smokers (ND)</li> <li>Healthy and diseased sites in smokers (SH+SD): significantly less IL-8, IP-10, MCP-1, MIP-1a, RANTES compared with healthy and diseased sites in non smokers (NH+ND)</li> <li>No significant differences either in healthy or in diseased sites between smokers and non smokers concerning IL-2, IFN-γ, IL-3, IL-4</li> <li>Diseased sites in smokers (SD): significantly less IL-7, IL-15 compared with diseased sites in non smokers (ND)</li> </ul>
Anil et al. 2013	90 subjects <ul style="list-style-type: none"> <li>30 periodontally healthy controls</li> <li>30 smokers with periodontitis</li> <li>30 non smokers with periodontitis</li> </ul>	Smokers: ≥20 cigarettes/day ≥2 years Non smokers: never smoked	Microcapillary pipette at the entrance of the gingival sulcus	MCP-1		MCP-1: significantly higher in smokers

Petropoulos et al. [23] for IL-1a. Additionally, Tumkiw et al. [24] found decreased IL-7 and IL-15 in the smoking subjects.

Fraser et al. [25] showed decreased level of soluble intercellular adhesion molecule -1 (sICAM-1) in smokers, Rai et al. [26] higher levels of myeloperoxidase (GM), and Anil et al. [27] higher levels of monocyte chemoattractant protein-1 (MCP-1).

Buduneli et al. [28] evaluated the effect of smoking on the Plasminogen Activator (PA) system proteins in GCF and serum of patients and compared healthy and diseased sites. The parameters studied were the tissue/blood vessel type PA (t-PA), urokinase type PA (u-PA) and inhibitors of the PA system (PAI-1 and PAI-2). Significant differences among smokers and non-smokers were found only for u-PA. They also studied various other biological parameters in GCF and found no effect of smoking [29]. Kurtis et al. [30,31] assessed many parameters among smokers and non-smokers and the only significant difference they found was decreased levels of Thiobarbituric Acid Reactive Substances (TBARS) in smokers who received supportive periodontal therapy combined with flurbiprofen compared to non-smokers who had the same intervention.

## Discussion

The present review assessed all current (up to May 2014) available evidence about the correlation of smoking and the composition of GCF in patients with chronic periodontitis. The majority of the reviewed articles studied the differences

in the GCF levels of various cytokines and other inflammatory mediators among smokers and non-smokers. It is evident that there are great differences among the relevant studies, which makes comparison impossible. It is also noteworthy that only one study [25] measured the levels of cotinine in GCF and plasma to confirm the smoking status of the patients.

With regards to interleukins, the results are conflicting. The existing articles study many different types of interleukins, so an overall conclusion cannot be drawn. The interleukins mostly studied were IL-1a, IL-1β and IL-1ra, for which some authors found differences between smokers and non-smokers and others showed no difference. Some studies investigated other biological parameters, such as IL-6, TNF-a, sICAM-1, IL-7, IL-15 and TBARS, but since there are no similar studies for comparison, their results cannot be yet of important value.

Finally, three studies utilized a different point of view, investigating whether there is any difference in the composition of GCF in smokers and non-smokers, before and after the periodontal therapy. Among the various parameters studied, significant differences were noticed after therapy only in smoking patients receiving flurbiprofen in combination with periodontal treatment.

## Conclusion

The existing literature cannot provide a solid conclusion with regards to the effect of smoking in the composition of GCF fluid in periodontal patients. Further studies of similar methodology and focus of investigation are needed, in order

to determine the precise relationship between smoking and the GCF profile in patients with chronic periodontitis.

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