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TLR4 and IL-18 gene polymorphisms in rheumatoid arthritis and periodontitis

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Introduction

Recent literature has indicated that patients with rheumatoid arthritis (RA) are at higher risk of developing moderate to severe periodontitis (PA) than non-RA patients. Several biologically plausible causal and non-causal mechanisms might account for this association between periodontitis and RA.
Bacterial components or virulence factors are involved in modulating inflammatory responses of periodontal tissue (Madianos et al. 2005). Host cell receptors recognize these bacterial components, thus triggering signaling pathways which lead to inflammatory responses and to the development of innate and acquired immunity. Among the receptors that recognize and bind pathogen-associated molecular patterns (PAMPs), Toll-like receptors (TLR) act as key-receptors of the innate immune system. In RA, the innate immune system is also centrally involved in articular pathophysiology via Toll-like receptor signaling by macrophages, dendritic cells, and fibroblasts, which subsequently leads to the early recruitment of inflammatory cells. One of the TLR4 dependent cytokines is interleukin-18 (IL-18). Depending on the immunological context, IL-18 has the capacity to induce either Th1 or Th2 cells in response to gram negative infections, and it is widely accepted that the balance of Th1 cells and Th2 cells is crucial to the immunoregulation in periodontitis. On the other hand, the importance of IL18 has been shown in the induction and perpetuation of chronic inflammation during experimental and clinical rheumatoid synovitis.
The role of IL-18 in periodontitis, as well as the function of TLR4 in cytokine activation imply that both genes coding for these proteins are candidate genes for susceptibility of both diseases: periodontitis as well as rheumatoid arthritis.

Objectives

In both diseases, PA and RA, genetic variants influence inflammatory immune responses, and polymorphisms of the IL-18 and TLR4 genes could play a role. Therefore, the aim of the study was to evaluated known polymorphisms of these two genes in RA patients and healthy individuals with or without periodontitis.

Material and Methods

68 RA patients and 203 non-RA control subjects volunteered to participate in this case-control association study. Clinical periodontal examination included probing depth (PD) and clinical attachment level (CAL). The presence of periodontitis was defined as proposed by the CDC Periodontal Disease Surveillance Workgroup (Page and Eke, 2007).
DNA was purified from peripheral blood leukocytes of all subjects using the QiaAmp blood DNA purification kit (Qiagen, Hilden, Germany). DNA samples were amplified by PCR using specific primers and protocols for each examined polymorphism as described previously (Folwaczny et al. 2004, Folwaczny et al. 2005, Noack et al. 2008). In the IL-18 promoter region, SNPs at position c.-838C>A and c.-368G>C from the translation start side were selected for genotyping representing two main IL-18 promoter haplotypes. Two known SNPs in the extracellular domain of the TLR4 gene (c.896A>G, p.Asp299Gly and c.1196C>T, p.Thr399Ile) were genotyped after amplification of TLR4 exon 3.
Allele frequencies, genotype distributions, and carrier frequencies of the variants in the study groups were calculated. Genotype distributions were compared between the RA group and the non-RA control group as well as between periodontally healthy and periodontitis controls (Fisher's exact test). A potential association between RA and the analyzed gene variants was assessed by using multivariat logistic regression models after adjusting for other confounding factors (gender and age). In a second regression model, the risk of the occurrence of both RA and severe periodontitis together was analyzed depending on the carriage of gene variants.

Results

The frequency of RA patients suffering from periodontitis was high: moderate periodontitis 41.2% (28 patients out of 68), and 48.5% (n=33) RA patients exhibited a severe periodontitis. The genotype and allele frequencies of the two IL-18 promoter variants were not statistically different between the RA patients and healthy individuals (figure 1,2). In contrast, TLR4 variants occurred in complete linkage disequilibrium, and were associated with RA, with an increased carrier rate of the c.896G and c.1196T allele in the RA patients (fig.3: 25% versus 9.9% in controls, P < 0.002, chi square test; adjusted odds ratio [OR] for age and gender: 2.29; 95% confidence interval [CI]: 1.07-4.92). There was no significant association between any of the analyzed gene variants and periodontal status in the control group. However, carriage of a TLR4 c.896G/c.1196T allele increased the risk of the occurrence of both RA and severe periodontitis together with an OR adjusted for sex, age and smoking status of 3.67; 95% CI: 1.5-8.9, (table).

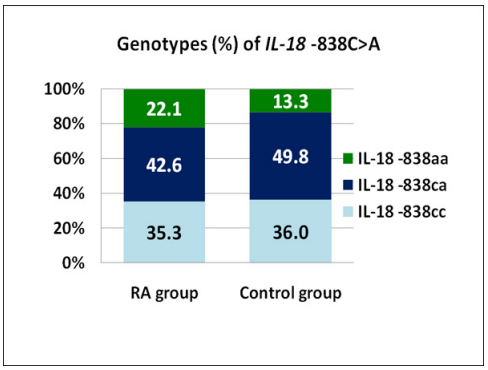

Fig. 1

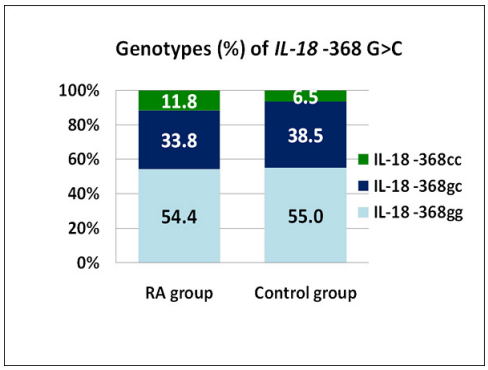

Fig. 2

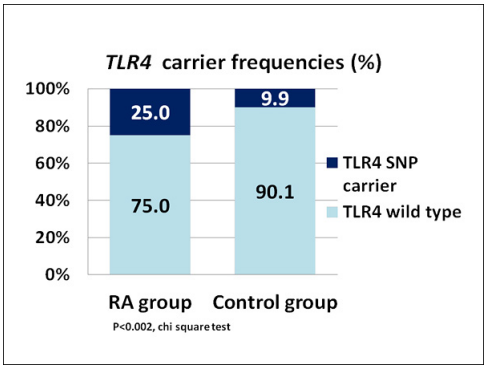

Fig. 3

Genotype	RA and no or moderate PA N (%)	RA + severe PA N (%)	Controls (severe PA, no RA) N (%)	Healthy controls (no RA, no PA) N (%)
TLR4 c.896AA/c.1196CC	27 (80.0)	23 (69.7)	116 (91.3)	50 (87.7)
TLR4 c.896AG/c.1196CT	7 (20.0)	10 (30.3)	11 (8.7)	7 (12.3)

P=0.035 RA+PA vs. healthy controls (chi square test)
P=0.001 RA+PA vs. PA (chi square test)

Tab. 1

Conclusions

TLR4 gene variants could contribute to the susceptibility to developing RA and to developing periodontitis in RA patients. These findings support the hypothesis of a shared genetic background between RA and periodontitis.

Financial support for the study was provided by the German Society of Dentistry (Düsseldorf, Germany)

Literature

Page RC, Eke PI. Case definitions for use in population-based surveillance of periodontitis. J Periodontol 2007; 78:1387 1399.

This Poster was submitted by Priv. Doz. Dr. Barbara Noack.

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Poster Faksimile:

TLR4 and *IL-18* Gene Polymorphisms in Rheumatoid Arthritis and Periodontitis

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4356

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Aim of the study

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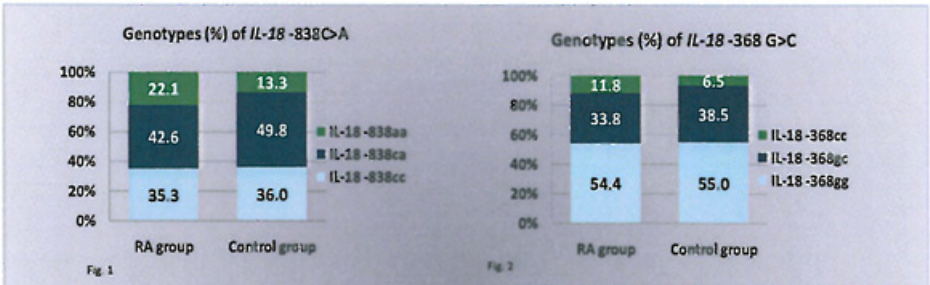
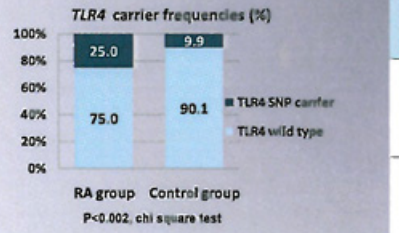


Fig. 1

Fig. 2



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