GENOTYPIC AND PHENOTYPIC CHARACTERIZATION OF PORPHYROMONAS GINGIVALIS IN RELATION TO VIRULENCE

AKADEMISK AVHANDLING

Som för avläggande av odontologie doktorsexamen vid Göteborgs Universitet kommer att offentligen försvaras i föreläsningssal 3, Instutionen för Odontologi, fredagen den 9 mars 2007 kl.13.00

Av

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Avhandlingen är av sammanläggningstyp och baseras på följande delarbeten:

Ι	Dahlén G, Gmur R, Yoshino T . Phenotypes, serotypes and antibiotic susceptibility of Swedish <i>Porphyromonas gingivalis</i> isolates from periodontitis and periodontal abscesses. Oral Microbiol Immunol 2007;21 (in press).
II	Yoshino T, Laine ML, van Winkelhoff AJ, Dahlén G. Genotypic characterization of <i>Porphyromonas gingivalis</i> isolated from Swedish patients with periodontitis and periodontal abscesses. Oral Microbiol Immunol 2007;21 (in press).
III	Yoshino T, Laine ML, van Winkelhoff AJ, Dahlén G . Genotypic variation and capsular serotypes of <i>Porphyromonas gingivalis</i> from chronic periodontitis and periodontal abscesses. FEMS Microbiol Lett 2007 (in press).
IV	Yoshino T, Björkner A, Dahlén G. Interaction of <i>Porphyromonas gingivalis</i> with human epithelial cells. Manuscript.

Genotypic and phenotypic characterization of *Porphyromonas* gingivalis in relation to virulence

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ABSTRACT

The present thesis was designed to increase the knowledge on the role of pathogenic potential of *Porphyromonas gingivalis* as a putative periodontal pathogen. *P. gingivalis* was selected to be the model species for periodontal pathogens based on expressing a number of significance virulence factors and on the considerable genetic heterogeneity of this species. The hypothesis of the present studies was that the pathogenic potential of *P. gingivalis* differs among this species and that certain clonal type of *P. gingivalis* express more pathogenic capacity than the others. The over all aim of this thesis was to investigate the virulence properties and the pathogenic capacity of species *Porphyromonas gingivalis*.

• Phenotypic heterogeneity of *P. gingivalis* species was evaluated by colony morphology, biochemical tests, enzymatic profiles, gas-liqid chromatography, antibiotic tests, SDS-PAGE profiling for cell wall protein and serotyping by monoclonal antibodies reactivity (study I).

• The diversity of whole chromosomal DNA among *P. gingivalis* species was evaluated by using amplified fragment length polymorphism (AFLP) and random amplified polymorphic DNA (RAPD) genotyping assays (study II).

• The variations of specific virulence biotypes based on *fimA*, *rgpA* and *kgp* genes and capsular K-antigens in *P*. *gingivalis* species were evaluated (study III).

• The interaction of *P. gingivalis* species with epithelial and PMN cells was evaluated by KB epithelial cell binding assay (study IV).

The present thesis demonstrated that:

- P. gingivalis strains showed a strong homogeneity in relation to biochemical tests and antibiotic susceptibility. Furthermore, the majority of P. gingivalis strains displayed monoclonal antibodies (MAbs) serotype A, while serotype B was unusual among P. gingivalis strains (Study I).
- P. gingivalis isolated from Swedish subjects with periodontitis and periodontal abscess exhibited a wide variety of genotypes with weak clustering pattern. No predominant genotype at the whole chromosomal DNA level was present among these P. gingivalis (Study II).
- Chronic periodontitis is not associated with a particularly virulent genotype of *P. gingivalis*. A highly virulent genotype (e.g. strain W83) of *P. gingigvalis* can be detected in certain periodontitis subjects (Study III)
- All strains showed binding capacity to host epithelial cells. Encapsulated *P. gingivalis* compared to nonencapsulated strains displayed a significantly lower binding capacity to host cells. No significant difference in binding and invasion was found between specific virulent genotypes. Thus, the two major virulence groups within *P. gingivalis* were mainly related to the presence/absence of a capsule structure of this organism (Study IV).

In conclusion:

P. gingivalis isolates from swedish periodontal disease cases express a considerable homogeneity in most phenotypic characteristics, although variations were found in colony morphology and MAbs and capsular antigen types. On the genotype level a considerable heterogeneity was found both at whole chromosomal level as for specific virulence genes. The studies support that there is generally a non-clonal structure of *P. gingivalis* although some specific virulent clones might be found infrequently in periodontitis. A capsule seems to be of particular important for *P. gingivalis* pathogenicity.

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