PERIODONTITIS AND PRETERM BIRTH
ORAL HEALTH STATUS OF PREGNANT WOMEN IN
SOUTH-EAST HUNGARY

Ph.D. Thesis
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SUMMARY

Relating the raising questions in the periodontal medicine in the last 15 years a case control study was undertaken to investigate that initial localized chronic periodontitis could be a risk factor for preterm birth and fetal growth restriction. The aim of the study was also to assess the overall oral health status, including caries and periodontal status, and microbial flora in the gingival crevice, of pregnant women in South-East Hungary, and to reveal the effect of demographic and socioeconomic status on this state.

Material and methods: Healthy, otherwise non-selected postpartum women were included into the study. A preterm birth (PB) case was defined if a patient had a threatening premature event during pregnancy, preterm premature rupture of membranes, or spontaneous preterm delivery, before the 37th week of pregnancy, and/or the weight of the newborn was less than 2500 g. Into the case group, 77 women were allocated, while 84 women belonged to the control group, all of whom had delivery after the 37th gestational week and with a newborn weighing ≥ 2500g. Demographic, socioeconomic status and known risk factors, like smoking, alcohol consumption were recorded. Dental examinations were performed in accordance with the WHO guidelines in the Department of Obstetrics and Gynecology, University Szeged. Microbiological samples were taken from the three deepest pockets, and culturing according to appropriate protocol.

Results: A significant association was found between PB and initial localized chronic periodontitis, the criteria being bleeding at ≥ 50% of the examined sites and having at least at one site at ≥ 4mm probing depth (P<0.001). The average weight of newborns of mothers with periodontitis was significantly less than in the women without periodontitis (P=0.002). The DMFT index for the women examined was 12.45. Parameters of the periodontal status were the follows: mean plaque index 0.67, frequency of calculus 21.07%, mean probing depth 1.67mm and frequency of bleeding on probing 37.80%. The periodontal status was worse in the case group, but the difference relating
the periodontal parameters was not significant. Caries and periodontal status were influenced by demographic and socioeconomic status. Microbial findings showed a significance difference between the case and control group. The result of the multivariate logistic regression analysis was an adjusted odds ratio for initial localized chronic periodontitis was 3.3157, 95% CI: 1.6418 - 6.6963, i.e. the second most important factor for preterm birth was the initial localized chronic periodontitis of the mother. The other most important influencing factor of preterm birth in this study proved to be smoking, with an adjusted odds ratio 4.5495, 95% CI: 1.2037 - 17.1955.

**Conclusions:** The results support the hypothesis that initial localized chronic periodontitis of pregnant women could lead to adverse pregnancy outcome and birth weight reduction.

### 1. INTRODUCTION

Recently the concept of periodontal “focal infection” has re-emerged, based on new data that suggest that periodontal infections may contribute to the morbidity and mortality of certain systemic conditions. Hence research demonstrates that oral diseases are not only markers of underlying health problems but also important determinants influencing the development of adverse chronic health conditions.

Concerning the proposed association between periodontal infection and preterm birth and low birth weight, which is still a significant public health problem and a major cause of neonatal death, it was decided to examine if this hypothesis was sustainable in a Middle-European Caucasian population.

#### 1.1. Epidemiology of preterm birth

Preterm birth is of considerable problem even in economically developed countries, as an estimated 11% of pregnancies end in preterm birth. In Hungary, the number of live births per year is around 95,000, of which about 8,500 (8.9%) are preterm
deliveries according to data of the National Institute of Obstetrics and Gynecology from 2000-2004, with the frequency of low birth weight infants being about 8.7%. These numbers have not decreased in the last 20-30 years.

Preterm birth and infant mortality results in many problems for the family involved, as well as for society in general. The societal burden of PLBW can be measured in terms of both morbidity and economic outcomes.

2. AIMS OF THE STUDY:

1. To study the possibility of any association between the periodontal status of pregnant women and preterm birth subjects in Hungary.
2. To examine whether the initial periodontitis could be an independent risk factor for preterm birth in Hungary.
3. To reveal if the periodontal status of pregnant women could influence the birth-weight of infants.
4. To find if any characteristic parameters of periodontitis can correlate with the incidence of preterm birth or low birth weight.
5. To assess the caries and periodontal status of pregnant women in a sample from South-East Hungary.
6. To examine the effects of socioeconomic factors on the caries status of a sample of pregnant women in South-East Hungary.
7. To evaluate the effects of socioeconomic factors on periodontal status of a sample of pregnant women in South-East Hungary.
8. To compare the caries status of pregnant women with previous data from Hungary, to determine if any improvement has occurred in the last 30 years.
9. To determine if there are any differences between the composition of the subgingival microflora of pregnant women with preterm birth comparing to those of normal delivery.
3. MATERIAL AND METHODS

3.1. Population sample

In total, 161 women were included in this case-control study. The patients participating in the study were volunteers who had delivery in the Department of Obstetrics & Gynecology, University of Szeged. They were informed about the aim of the study and a detailed Ethics Committee Approved Consent Form for dental, obstetrical and microbiological investigation was signed by them. Only systemically healthy women, carrying a single fetus, without any sign of inflammation, were enrolled into the case-control study. Each examination was completed within three days post-partum by the dental member of the team, who was blind as to case-control status.

3.2. Collecting demographic and socioeconomic data

All subjects had a record detailing personal data, medical and obstetrical history, including the number of previous pregnancies, as well as to prenatal care. Before the dental examination a questionnaire, which included queries regarding the socio-economic status of both the patient and the father of the child, was completed by each subject after they received necessary instruction by the dentist. Adverse habits e.g. smoking, alcohol- or drug-abuse were also sought. Regarding smoking, alcohol and/or drug consumption, there was a choice between ‘Yes’ or ‘No’ answers. However, details regarding the quantities consumed (or their frequencies) were not gone into.

The socio-economic status was pursued and categorized by educational level, occupation, and residence area of the patient and father. The level of education was divided into four categories i.e. primary school (8 years), technical school (3 yrs post-primary school), grammar-school (4 yrs post-primary school), and higher education (university or college), as were three types of occupation i.e. manual workers, intellectuals/professionals or ‘other occupations’ (e.g. shop assistant, housewife,
unemployed person, etc.). Regarding their place of residence, patients were divided into urban and rural groups.

3.3. Caries and periodontal status measurements

The caries examinations were performed following WHO guidelines in the Department of Obstetrics & Gynecology, where the subjects were seated in a comfortable chair with a head support, and a dental light-source was used to standardize the examination conditions. Third molar teeth were excluded. DMFT (Decayed, Missing, Filled Teeth) and DMFS (Decayed, Missing, Filled Surfaces) indices were calculated, with retained roots and crowned teeth regarded as carious and/or filled surfaces respectively.

A detailed periodontal status was recorded, which evaluated plaque, calculus, recession, tooth mobility, probing depth and bleeding on probing. The plaque index was registered according to the criteria of Silness & Löe on the “Ramfjord teeth”, at four surfaces per tooth. The presence or absence of supragingival calculus was recorded dichotomously on all the teeth. Buccal marginal gingiva recession was measured from the cemento-enamel junction to the marginal gingival, and recorded in mm. Tooth mobility was assessed on a 0-3 scale. Probing depth was measured from the gingival margin to the most apical penetration of the probe at six sites per every tooth. Bleeding on probing (BOP) was regarded positive if it occurred within 15 seconds after measuring the probing depth at the teeth, and was recorded dichotomously.

All caries and periodontal data were analyzed for significant relationships with respect to the age, educational level, profession, and place of residence, number of children and regarding preterm birth and low birth-weight.

3.4. Assessment of newborns’ weight

Newborns were weighed immediately after delivery on a precise scale, which is regularly used. Mothers according to the weight of the newborns were allocated to the case or control groups (More detailed see 3.6).
3.5. Microbiologic assessment

**Microbiological sampling:** The area to be sampled was isolated with cotton rolls; the tooth surface was cleaned with 70% ethanol and dried with sterile cotton swabs. Samples were obtained from the three deepest pockets or most diseased sites with individual sterile paper points, which were placed in the gingival crevice for 15 s, all cultures were commenced within 1 h.

**Sample culturing:** The samples were placed into 1.0 ml reduced BHI and homogenized. Then the suspensions were diluted ($10^{-1}$-$10^{-6}$) in reduced BHI (Brain Heart Infusion) broth, and 100 µl samples of each dilution and 100 µl samples of the corresponding undiluted suspension were immediately plated on selective and non-selective media. Columbia agar base supplemented with 5% (v/v) cattle blood, haemin and vitamin K₁ was used to quantify the total cultivable facultative and anaerobic bacterial flora. *Veillonella* spp. were isolated from Veillonella agar, and Rogosa agar was used for the selective isolation of lactobacilli, CFAT agar for *Actinomyces* spp., and chocolate agar for determination of the total aerobic bacterial flora. For the selective growing of streptococci and *Enterobacteriaceae*, Mitis Salivarius and Endo agar, respectively, were used. For aerobic bacteria, the plates were cultured at 37 °C in a 5% CO₂-containing environment for 48 h. For the isolation of anaerobic organisms, cultures were set up and incubated in an anaerobic chamber for 6 days at 37 °C. Each different colony type from positive cultures was subcultured for purity and identification. API 20A and ATB ID 32 ANA tests were used to identify anaerobic bacteria, facultative anaerobic Gram-positive cocci and bacilli. Other conventional tests for different bacteria were used where appropriate.
3.6. Case definition

Mothers fell into the case group if the newborn’s weight was less than 2500g, or the mother had spontaneous preterm labor or preterm premature rupture of membranes before the 37th gestational week. Women, who had threatening preterm labor, were treated by a special protocol to maintain their pregnancy for as long as possible. As a result, in some cases labor was delayed until week 37, and these mothers were then allocated to the case group. Mothers, whose delivery occurred after week 37 without any previous problem, and had a newborn with a weight ≥2500 g, were placed in the control group.

3.7. Criteria of initial localized chronic periodontitis

The “critical probing depth” was used to categorize a mother with periodontal disease. A woman had periodontitis if she had ≥4 mm probing depth at least at one site and bleeding on probing at ≥50 % of teeth. These criteria for initial localized chronic periodontitis were chosen because a 4mm pocket depth represents a disease level, when there is always an attachment loss. Bleeding shows that the periodontitis is in an active phase. Subjects, having no ≥4 mm pockets, or BOP occurring at less than 50 % of teeth, were regarded as periodontal healthy.

3.8. Statistical analysis

For the comparison of mean values, the t-test and one-way analysis of variance were used, as well as the Mann-Whitney and Kruskal-Wallis tests in the case of demonstrated non-normality. The normal distribution of samples was tested using the Kolmogorov-Smirnov test. The Spearman correlation coefficient was employed to assess correlations between continuous and ordinal variables. Categorical data were analyzed using the chi-square and Fisher’s exact test.

For the analysis of multivariable dependencies we used multiple linear regression analyses, as well as logistic regression for the calculation of adjusted odds ratios, with stepwise model selection, based on the likelihood ratio criterion.
4. RESULTS

4.1. Demographic and socio-economic data

Caries and periodontal data, and the date of delivery and birth weight of the infants of 161 women, were analyzed statistically. The mean age was 27.7 years, and the age distribution was found to be normal, with no significant difference between the mean age of the case and control groups (P=0.589).

4.2. Caries status of postpartum women

The DMFT index for the whole population was 12.45, while the DMFS was 26.07. The average number of decayed teeth was 1.94, filled teeth 6.29, and the mean for missing teeth was as high as 2.86. Women with higher educational level and intellectuals had significantly less caries, radices and more fillings. The number of decayed teeth increased if the women had more children, but the difference was not significant.

The result of the regression analysis showed that the DMFT index was influenced mostly by the mother’s age (P<0.001) and amount of plaque (P=0.004), while the number of pregnancies was not amongst the most important factors (P=0.115). In comparing the current results with former Hungarian caries values, there is no sign of any improvement in caries frequency during the past thirty years (DMFT 12.45 Orosz et al. 1975; 12.5 Bánóczy et al.1978; 13.68 Orosz et al. 1980; 12.93 Papp et al. 1990).

4.3. Periodontal status of postpartum women

The mean plaque index was 0.67 (case: 0.72, control: 0.62), frequency of calculus 21.07% (case: 23.35, control: 18.98%), mean probing depth 1.67mm (case: 1.72mm, control: 1.62mm) and bleeding on probing occurred at 37.80% of the teeth (case: 43.29%, control: 32.76%). The periodontal status was worse in the case group, but the difference was not significant. The existing of probing depth ≥4 mm at least at one site and bleeding
on probing at $\geq 50\%$ of the teeth at the same time, had a significant difference between the case and control group ($P<0.001$).

According to the result of the regression analyses each periodontal score was related to educational level ($P<0.001$). The frequency of calculus, probing depth and frequency of bleeding on probing were also influenced significantly by the age of the patients.

4.4. Microbiological results

Anaerobic bacteria could be detected in higher numbers (average colony-forming units/ml, CFU/ml) in all of the subgingival plaque samples from the patients in case and control groups with the clinical evidence of initial periodontitis. The average number of isolated anaerobic species was 8-14/patient with periodontitis, and 0-4/patient without the clinical evidence of periodontitis. The bacterial numbers associated with periodontitis were up to $10^5$ times higher than those associated with the patients without periodontitis.

The anaerobic bacterial culture positivity showed differences between the case and control groups, but no differences were seen in the prevalence and distribution of aerobic bacteria.

4.5. Association between periodontal infection of mother and preterm birth

The results of preliminary univariate analyses the analysis showed that having $\geq 4$ mm probing depth at least at one site and bleeding on probing at $\geq 50\%$ of the teeth, had a significant relationship with preterm birth in this study group ($P<0.001$). The total number of mothers with initial localized chronic periodontitis was 57, among them 39 (68.4%) women being in the case group and only 18 (31.6%) in the controls, the difference was significant ($P<0.001$).

The mean weight of periodontitis mothers’ newborns was 2834.5g, while in the control group it was higher at 3180.3g; the difference was significant ($P=0.002$).

The adjusted odds ratio for initial localized chronic periodontitis was 3.3157, 95% CI: 1.6418 - 6.6963, i.e. the second most important factor for preterm birth was the initial
localized chronic periodontitis of the mother. The other most important influencing factor of preterm birth in this study proved to be smoking (only 17 women admitted smoking during pregnancy), with an adjusted odds ratio 4.5495, 95% CI: 1.2037 - 17.1955.

5. DISCUSSION

The subject group in this study was representative of the population from which the sample was taken. Relating caries status of pregnant women, there is no sign of any improvement in caries frequency during the past thirty years. The most important influencing factors on caries status were the age of the women and the plaque index, and not the number of previous pregnancies. The periodontal status of mothers was better if they were highly educated, worked as intellectuals, lived in cities, were younger, and had fewer children. The results of the microbiological examinations demonstrated that the number of anaerobic flora (including the key pathogens of periodontitis) was significantly higher, and the composition of the flora was more complex, among those women associated with PB and/or a lower birth weight than in the control group.

Results of the study confirm a significant association between initial localized chronic periodontitis of the patient during pregnancy and preterm birth, as well as low birth weight.

Periodontal infection is an easily detectable and readily treatable illness. Its treatment costs are very low as compared to the prenatal treatment of women with threatening preterm birth, and the perinatal care-costs associated with that of a low birth weight infant. Using the results of applied “periodontal medicine”, the likelihood of systemic disease (amongst which is preterm delivery), can be reduced and patients’ lives significantly improved.
6. CONCLUSIONS

1. We found an association between periodontitis of pregnant women and preterm birth in the study group in Hungary.

2. We found that already initial localized chronic periodontitis had a significant association between adverse pregnancy outcomes.

3. We found that in the group investigated, the initial localized periodontitis of mothers was associated with a significantly smaller birth weight of newborns.

4. We found that at least one $\geq 4$ mm probing depth pocket and bleeding on probing at $\geq 50\%$ of teeth influenced the birth weight and time of delivery.

5. We found that in the population investigated, the age and number of pregnancies influenced the caries status significantly, while educational level, the occupation and place of residence had no such influence. The age was the most important factor.

6. We found that in the population investigated, the status of the periodontium was influenced mostly by educational level and occupation, while the age, place of residence and the number of previous pregnancies had only a smaller or no effect.

7. We concluded that the DMFT index of pregnant women showed no signs of having improved in the previous 30 years in Hungary.

8. The study found microbial evidence that maternal periodontal disease and the presence of key pathogens are significant contributors to the obstetric risk of preterm delivery.

Following the found evidences, a dental surgery has been established in the Department of Obstetrics and Gynecology in Szeged, presumably first time in the world. Here professional oral health measurements for those in-patients, who are treated because of threatening premature labor and have gingivitis or periodontitis, are now regularly provided.
LIST OF SCIENTIFIC PUBLICATIONS RELATED TO THE THESIS:

1. Radnai M, Gorzó I. A fogágybetegség mint a koraszülés és a kis súlyú újszülött születésének lehetséges rizikó faktora. (Irodalmi áttekintés) Fogorvosi Sz. 95; (6) 241-244, 2002.


LIST OF CONGRESS PRESENTATIONS WITH ABSTRACT RELATED TO THE THESIS CAN BE CITED:

   *Caries Res.* 29; (4) 302 No 36, 1995

   *Fetal Diagn Ther.* 17 (Suppl 1) No 165, 2002

   Europerio 4, 19-21 June 2003, Berlin, Germany  
   *J Clin Periodontol.* 30; Suppl. 4, No 133, 2003


   *Fogorvosi Sz.* 97; (1) 50, 2004.

6. **M. Radnai**. Preventive Program for Improving the Oral Health of Women in Hungary. P4  
   ADEA American Dental Education Association 2nd International Women’s Leadership Conference June 20-23, 2003, Göteborg, Sweden  
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7. Urbán E, Nagy E, **Radnai M**, Gorzó I, Novák T, Pál A. Microbial investigation of the possible association between pre-term birth and early periodontitis.  
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