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Changes in inflammatory mediators in gingival crevicular fluid following periodontal disease treatment in pregnancy: relationship to adverse pregnancy outcome

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ABSTRACT

Periodontal disease (PD) in pregnancy is associated with an increased risk of adverse pregnancy outcomes including miscarriage and preterm birth. Evidence exists that periodontal disease treatment may reduce inflammatory mediators in gingival crevicular fluid (GCF) and the risk of inflammation-associated pregnancy complications. The aim was to determine if periodontal disease treatment during mid-pregnancy alters local inflammation in GCF and has beneficial effects on clinical dental parameters. Eighty pregnant women with clinically diagnosed PD were recruited from a randomised controlled trial on the treatment of periodontal disease in pregnancy conducted in Perth, Australia. The treatment group underwent intensive PD treatment (20–28 weeks' GA), while the control group underwent the same treatment postnatally. GCF was collected at 20 and 28 weeks' gestation and concentrations of cytokines determined by multiplex assay: IL-1β, IL-6, IL-8, IL-10, IL-12p70, IL-17, TNF-α and MCP-1. Periodontal treatment significantly reduced the GCF levels of IL-1β, IL-10, IL-12p70 and IL-6 at 28 weeks' GA compared with controls, while levels of MCP-1, IL-8 and TNF-α exhibited a significant gestational age-dependent increase, but no treatment response. Post-treatment clinical parameters improved with significant reductions in bleeding on probing, clinical attachment loss, and probing depth. No changes in pregnancy-related outcomes were observed, although the severity of periodontal disease was significantly associated with an increased risk of infants born small for gestational age. PD treatment in pregnancy reduces the levels of some inflammatory mediators in the GCF and improves dental parameters, with no overt effects on pregnancy outcome.

1. Introduction

Periodontitis is an infectious disease process that, in its progressive stage, is a substantial contributor to global oral health disease. Approximately 10–15% of the adult population worldwide are affected by periodontal disease, with higher rates observed in indigenous populations (Petersen and Ogawa, 2005; Jamieson et al., 2010). Periodontal disease is characterised by destruction of the periodontal ligament, bone and soft tissue as a result of a local host immune response to microbial plaque on the tooth surface. If left untreated, this inflammation leads to alveolar bone resorption and, eventually, tooth loss (Kornman et al., 1997; Kinane, 2001). The severity and extent of the disease is modified by genetics, host

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response and lifestyle factors such as oral healthcare, smoking, age, race, hormonal changes, obesity, and diabetes (Kornman et al., 1997; Kinane and Chestnutt, 2000; Van Dyke and Sheilesh, 2005; Carrillo-De-Albornoz et al., 2010; Pretzl et al., 2012). The presence of periodontitis has been linked to systemic diseases beyond the periodontium, such as diabetes mellitus, cardiovascular disease, osteoporosis, pulmonary disease, and rheumatoid arthritis (Scannapieco, 2005; Otomo-Corgel et al., 2012).

Periodontal disease in pregnant women has been associated with an increased relative risk of adverse pregnancy outcomes such as late miscarriage, preterm birth, and delivery of low birth weight infants (Jeffcoat et al., 2001; Moore et al., 2004; Jarjoura et al., 2005; Offenbacher et al., 2006a; Shub et al., 2006). The prevalence and incidence of gingival inflammatory symptoms and periodontal disease are significantly higher in pregnant women and are associated with gestational age, obesity, smoking, racial origin, educational level, employment status and maternal age (Yalcin et al., 2002; Taani et al., 2003; Lieff et al., 2004; Vogt et al., 2012). Translocation of periodontal pathogens, pro-inflammatory cytokines and prostaglandins from the oral cavity to the uterus and feto-placental unit provides a plausible biological mechanism for the proposed association between the periodontal disease and adverse pregnancy outcomes (Han et al., 2004).

Gingival crevicular fluid (GCF) collected from the gingival crevice surrounding the teeth is composed of cellular exudates from microbial plaque, immune cells, and resident periodontal tissues. Measurement of inflammatory mediators in GCF can be used to determine the current periodontal inflammatory status, while clinical dental parameters are more indicative of long-term periodontal disease progression (Embery and Waddington, 1994). There is ample evidence demonstrating that the treatment of periodontal disease improves the inflammatory profile of GCF, decreasing the levels of a variety of cytokines such as IL-1β, IL-6, IL-8, IL-10, IL-12, IL-17, TNF-α, chemokines (MCP-1, RANTES) and prostaglandins in the affected sites (Gamonal et al., 2000; Thunell et al., 2010; De Lima Oliveira et al., 2012; Gupta et al., 2013). Treatment and subsequent reduction of inflammatory mediator levels is also associated with significant improvements in the clinical parameters of the disease, such as probing depths, percentage of bleeding on probing and plaque (Ide et al., 2003; Offenbacher et al., 2006b; Fiorini et al., 2013); however, the effects of pregnancy on GCF inflammatory status in periodontal disease are not known. Pregnancy is a state of selective immune modulation, characterised by a decreased Th1-type response to some antigens and an increased Th2 response to others (Mor et al., 2011). Hence, the inflammatory profile of GCF in pregnancy and its response to treatment may be atypical and explain the risk of inflammation-associated pregnancy complications such as preterm birth, low birth weight and fetal growth restriction. Only one previous study has been published in this area: Fiorini et al. (2013) observed major reductions in GCF levels of IL-1β and IL-8 and periodontal inflammation after non-surgical periodontal therapy during pregnancy, with no significant effect on cytokine levels in the maternal circulation. The effects of treatment on pregnancy outcome were not investigated.

The aim of this study was to investigate the effect of periodontal disease treatment in mid-pregnancy on the inflammatory mediators and clinical dental parameters in women with clinically diagnosed periodontitis, and to assess the influence of gestational age on these parameters. A secondary aim was to determine if dental parameters and/or inflammatory mediator profile in GCF can be predictive of common adverse pregnancy outcomes such as preterm birth (PTB), low birth weight (LBW) and small for gestational age (SGA).

2. Materials and methods

The study population for this project consisted of a subset of 80 pregnant women with clinically diagnosed periodontal disease who were recruited to a randomised controlled clinical trial to assess the ability of treatment of periodontal disease in pregnancy to reduce the rates of PTB (The Smile Study: NCT00133926). The trial was carried out in Perth, Western Australia, between 2006 and 2009. Over 3000 women were recruited with written informed consent in accordance with approval conditions from the local human research ethics committee (Newnham et al., 2009). The inclusion criteria for this study were: (1) women older than 16 years of age with singleton pregnancies between 12 and 20 weeks' gestational age (GA) who had no known fetal anomalies; (2) no maternal cardiac disease that would require antibiotic therapy for dental treatment; (3) presence of at least 20 natural teeth; (4) no administration of periodontal treatment during the current pregnancy before recruitment. Periodontal pocketing was required to be $\geq$3.5 mm deep at 25% of sites. Periodontal pocketing is used to define the presence of periodontal disease as it better represents the microbial challenge than clinical loss of attachment. Of the 80 women in the sub-study, $n = 40$ had been randomised to receive an intensive course of periodontal disease treatment between 20 and 28 weeks of gestational age (GA) (intervention group: Rx), while the remaining 40 women had been randomised to receive similar treatment postnatally (control group: Ctrl). Samples from one participant in the control group were unavailable, resulting in $n = 39$; three participants had GCF samples for only one time point (*i.e.* either 20 weeks' or 28 weeks' gestational age).

2.1. Oral examination

At 20 weeks' GA (baseline) women in both the treatment and control groups underwent comprehensive periodontal examination using an automated constant force Florida probe (Florida Probe Corporation, Gainesville, FL, USA). Dental parameters and the data were stored digitally on a computer. Blood samples for leucocyte gene expression studies and GCF were collected simultaneously (see below). Periodontal treatment consisted of three visits each lasting 1 h and involved non-surgical debridement of the sub- and supra-gingival plaque and removal of calculus and overhanging restoration adjustments. Comprehensive oral hygiene instructions were provided at each visit with