Gingival and Periodontal Changes in Patients Undergoing In vitro Fertilization Treatment: A Clinical Study

Abstract

Aims: The aim of this study was to investigate the effect of in vitro fertilization (IVF) treatment on different parameters of periodontal status. Settings and Design: This was a clinical observational study. Materials and Methods: One hundred and seventy-nine patients who underwent IVF treatment according to the standard IVF protocols were examined using the simplified oral hygiene, gingival index (GI), sulcus bleeding index (SBI), and determining the clinical attachment loss (CAL). A full-mouth examination except for the third molars was performed at 4 sites per tooth (mesiobuccal, distobuccal, mesiolingual, and distolingual). Periodontal evaluation was performed before infertility treatment, at the end of infertility treatment, and 14 days after embryo transfer. Statistical Analysis: The Kruskal–Wallis or Fisher’s tests were used to compare the median or mean values as appropriate. Results: The oral hygiene index simplified was 0.49, 0.32, and 0.37 at pretreatment, on the day of human chorionic gonadotropin (HCG) trigger, and on the day of the pregnancy test, respectively. The GI showed significant differences before and after treatment. The mean GI was 0.13 at pretreatment compared to 0.51 and 0.53 on the days of HCG trigger and of the pregnancy test, respectively. The same trend was seen for SBI. There were no differences in CAL among the three examinations. There was no difference between the two groups except for GI (0.71 vs. 0.48 for a positive pregnancy test vs. nonpregnancy, respectively). Conclusions: IVF medications and a superphysiological condition affect oral health, particularly gingival and periodontal statuses, and likely complicate the relationship between infertility, sex hormones, and infertility management. Large-scale studies are needed to confirm the effect of such treatment on oral health.

Keywords: In vitro fertilization, oral health, periodontal status, sex hormones

Introduction

Assisted reproductive technology, mainly in vitro fertilization (IVF), is the cornerstone of treatment options available for these couples. According to the Center for Disease Control and Prevention, 1.5–2.0 million cycles of IVF are performed worldwide every year.[1] IVF treatment protocols include superovulation status for the ovaries, this will lead to estradiol levels which starts low and rises to 1000–4000 pg/ml by the time of the human chorionic gonadotropin (HCG) injection, significantly elevated or rapidly rising serum estradiol concentrations is a superphysiological condition.[2] Clinically, a positive correlation between estradiol and progesterone serum levels and gingival index (GI) inflammation has been demonstrated in many clinical situations, including pregnancy, puberty, and endogenous use of hormones such as oral contraceptives or even during menstrual cycles.[2-6]

The relationship between infertility and periodontal disease has been investigated by many researchers, who found a correlation between periodontal diseases and risk of infertility both in female and male partners,[7,8] however, few studies have investigated the effect of IVF procedures and medications on periodontal status.[9-12]

This study was designed to investigate the effect of IVF treatment on different parameters of periodontal status.

Materials and Methods

A total of 372 patients underwent IVF treatment over a 1-year period (from May 2015 to May 2016) at Hope Fertility Clinic and Islamic Hospital IVF Centre – Amman – Jordan. Of these, 261 of them fulfilled the inclusion criteria for the study, of whom 228 women agreed to be enrolled...
in the study. Informed consent was obtained from all couples who agreed to be part of the study. The protocol was conducted in accordance with the Helsinki Declaration of 1975, as revised in 2008, and ethical approval was obtained from the local Hospital Ethics Committee.

All 228 women were initially examined; 196 women completed the IVF treatment, and 17 of whom opted to withdraw from the study after completing the IVF cycle, leaving 179 women as the final participating number who were enrolled in the study.

Our inclusion criteria were as follows: women could not have medical diseases, pretreatment hormone intake, or previous infertility treatment including previous IVF trials in the preceding 6 months; the presence of at least 20 teeth and all teeth groups (incisors, premolars, and molars); and the absence of extensive tooth restorations (involving no more than ⅔ of the crown). Exclusion criteria were as follows: smoking, use of any medications, or history of dental treatment during the previous 6 months.

The ovarian stimulation protocols that were used were either a long agonist protocol or a short antagonist protocol. The decision on which stimulation protocol to be used was dependent on the local infertility treatment protocol used in the centers. In the long agonist protocol, pretreatment with triptorelin (0.1 mg of Decapeptyl®, IPSEN) began between days 21 and 24 of the preceding cycle (midluteal phase). Ovarian stimulation began when pituitary downregulation was established (i.e., the beginning of the menstrual cycle or when serum estradiol was <50 pg/ml or <200 pmol/l after 2 weeks if no cycle began). When pituitary downregulation was established, the triptorelin treatment was continued until and including the day of HCG administration.

In the short antagonist protocol, 0.25 mg of Cetrotide®, Merck Serono (cetrorelix acetate for injection) was administered daily subcutaneously from day 6 of ovarian stimulation until and including the day of HCG administration.

For women in both treatment regimens, ovarian stimulation was initiated from day 2 of the cycle using combined human menopausal gonadotropin (HMG) Menogon®, Ferring Pharmaceutical of variable doses (150–450 IU) depending on the local protocol. The daily dose was adjusted and individualized for each patient based on the follicular growth as observed by ultrasonography. On the day of HCG administration, no treatment with HMG was administered.

HCG (Choriomon®, IPSA) (10,000 IU in 1 ml of saline) was administered when at least three follicles were ≥18 mm, as measured by ultrasound. Then, 36 h later, oocyte retrieval was performed followed by IVF or intracytoplasmic sperm injection. No more than four embryos were to be transferred at 2–5 days after oocyte retrieval. Progesterone for luteal support (400 mg of Cyclogest®) was initiated according to each center’s routine practice for 2 weeks from ovum selection and continued for another 2 months if pregnancy occurred.

Patients were evaluated for periodontal status by a single blinded examiner using a mouth mirror and a 15-mm conventional periodontal probe (Hu-Friedy). Dental examination included the simplified oral hygiene,[13] GI,[14] sulcus bleeding (SBI),[15] index, and clinical attachment loss (CAL).[16] A full-mouth examination excluding the third molars was performed at 4 sites per tooth (mesiobuccal, distobuccal, mesiolingual, and distolingual). Periodontal status was evaluated 3 times for all women: before beginning infertility treatment, at the end of infertility treatment (day of ovulation triggering), and 14 days after embryo transfer (day of the pregnancy test).

**Statistical analysis**

The Kruskal–Wallis or Fisher’s tests were used to compare the median or mean values for characteristics and outcomes related to the IVF treatment. Student’s t-test with Bonferroni correction or one-way ANOVA was used to compare two or more mean values of indices before and after IVF, respectively. Statistical analyses were conducted using the SPSS version 18 software package (PASW®, IBM, USA). The level of statistical significance of all comparisons was set at 5% (P ≤ 0.05).

**Results**

In total, 179 women were enrolled in the final analysis of the results. Figure 1 shows the number of patients ultimately enrolled in the study.

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**Figure 1: Flowchart of patient enrollment in the study**

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The mean oral hygiene index simplified was 0.49, 0.32, and 0.37 at pretreatment, on the day of HCG trigger, and on the day of the pregnancy test, respectively, with no observed statistically significant difference [Table 1].

The GI showed statistically significant differences when patients before treatment and after treatment were compared, both on the day of HCG trigger and on the day of the pregnancy test. The mean GI was 0.13 at pretreatment compared to 0.51 and 0.53 on the day of HCG trigger and the day of the pregnancy test, respectively [Table 1].

The same trend was seen when the sulcus bleeding index (SBI) was examined, with means of 0.19, 0.69, and 0.71 at the pretreatment, day of HCG trigger, and day of the pregnancy test, respectively [Table 1].

CAL showed no statistically significant difference among the three examinations, with means of 1.71, 1.47, and 1.49 at pretreatment, on the day of HCG trigger, and the day of the pregnancy test, respectively [Table 1].

Table 2 shows the periodontal status parameters for both groups: those who became pregnant and those who did not become pregnant. There was no statistically significant difference between the two groups except for GI (0.71 vs. 0.48) for positive pregnancy test patient’s versus nonpregnant patients, respectively.

When the effect of the ovarian induction protocols on periodontal status parameters was examined, there was no statistically significant difference between both protocols (long vs. short) [Table 3].

**Discussion**

It has been suggested that sex hormones are among many important factors affecting the development of periodontal disease.[17] In our study, we assessed the gingival inflammation and periodontal status of women undergoing IVF treatment and the effect of different protocols and hormones on these changes.

Compared to pretreatment assessment, periodontal and gingival assessment on the day of HCG trigger when estrogen levels are highest revealed that patients had significantly higher gingival inflammation and bleeding with a similar oral hygiene index [Table 1]. This effect was also demonstrated at the time of the pregnancy test when patients were exposed to super physiological levels of endogenous progesterone used for luteal phase support. These findings may be attributed to increased levels of estrogen and progesterone. Sex steroid hormones have been shown to directly and indirectly influence cellular proliferation, differentiation, and growth in target tissues, including keratinocytes and fibroblasts in the gingiva.[19] The effect of estradiol on premenopausal gingival fibroblasts was demonstrated to induce cellular proliferation and depress protein production in a laboratory setting. This cellular proliferation appears to be the result of a specific population of cells within the parent culture that responds to physiologic concentrations of estradiol.[19] On the other hand, estrogen reduces the production of proteins, both collagen and noncollagen by fibroblasts.[20] The effect of sex steroids extended to increase the rate of folate metabolism in oral mucosa, thereby inhibiting tissue repair.[21]

The effect of estrogen in our patients may have been enhanced by high levels of exogenous progesterone that were supplied as adjuvant therapy to support the luteal phase in IVF, as progesterone enhances the vascularity of the gingiva and other nonperiodontal intraoral tissues,[22] in addition to modulating inflammatory responses through different mechanisms including increases in the production of prostaglandins,[23] polymorphonuclear leukocytes and prostaglandin E2 in the gingival crevicular fluid (GCF),[24] and reducing glucocorticoid anti-inflammatory effects.[25]

However, the effect of sex steroids on the gingiva and periodontal status has been described by many other studies that demonstrated that endogenous sex steroid hormones play significant roles in modulating the periodontal tissue responses and may alter periodontal tissue responses to microbial plaque and directly contribute to periodontal disease. They can influence the periodontium at different life stages including puberty, menstruation, pregnancy, menopause, and postmenopause,[26] although only a four studies have examined the effect of IVF treatment on the gingiva and periodontal status.

Haytaç et al. demonstrated in their study the effect of combining gonadotropin injection therapy with other ovulation therapy on gingival inflammation (GI), bleeding on probing, and GCF volume and concluded that duration of the usage of these drugs is strongly associated with the

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**Table 1: Periodontal examination during in vitro fertilization treatment**

| Periodontal status parameters | Examination 1 (pre-IVF) Mean±SD | Examination 2 (ovulation trigger day) Mean±SD | Examination 3 (pregnancy test day) Mean±SD | E1 versus E2 | E1 versus E3 | E2 versus E3 |
|-------------------------------|----------------------------------|-----------------------------------------------|------------------------------------------|--------------|--------------|--------------|
| OHI-S                         | 0.49±0.031                       | 0.32±0.028                                    | 0.37±0.030                               | 0.1          | 0.2          | 0.2          |
| GI                            | 0.13±0.012                       | 0.51±0.032                                    | 0.53±0.042                               | 0.001*       | 0.001*       | 0.3          |
| SBI                           | 0.19±0.01                        | 0.69±0.043                                    | 0.71±0.044                               | 0.001*       | 0.001*       | 0.3          |
| CAL                           | 1.71±0.15                        | 1.47±0.18                                     | 1.49±0.15                                | 0.1          | 0.1                 | 0.3          |

*Statistical significance. OHI-S=Oral hygiene index simplified, GI=Gingival index, SBI=Sulcus bleeding index, CAL=Clinical attachment loss, SD=Standard deviation, IVF=In vitro fertilization
severity of gingival inflammation,\cite{Pavlatou2011} reflecting the role of super-physiological levels of ovulatory hormones on these indices.

Pavlatou et al.\cite{Pavlatou2016} concluded that periodontal clinical parameters worsened in women undergoing IVF treatment. However, they also studied the preexisting periodontal status and found that poor status appeared to be associated with poorer outcomes of IVF treatment.

Lalasa et al. concluded that infertility treatment exacerbates the gingival inflammation and periodontal disease process by reporting significantly higher gingival inflammation and SBI in IVF-treated women when compared to untreated women. Furthermore, they revealed that those suffering from infertility problems had statistically higher CAL compared with the fertile group. These findings correlate with our results with changes in the gingival sulcus bleeding indices.\cite{Lalasa2004} Another recent study demonstrated that IVF medications, both oral and injectable, have an effect on gingival inflammation reflected by more bleeding on probing and increase gingival fluid volume levels.\cite{Janssens2016}

The occurrence of pregnancy did not worsen the studied indices between those who become pregnant and those who did not; all parameters remained consistent (GI, SB, and CA). This finding can be explained by the fact that the nonpregnant group were still under the influence of super-physiological levels of both estrogen and progesterone at the time of the pregnancy test [Table 2].

When different IVF treatment protocols were compared, there was no difference in all parameters between the long protocol group and the short protocol group, indicating that there was no effect of the type of ovarian suppression on the periodontal and gingival status [Table 3].

In addition to the small number of women that were enrolled in the studied group, other limitations of our study include the lack of a control group, absence of testing of other cofactors which may affect the results, such as psychological factors, pre-IVF treatment, and other medical diseases associated with infertility that may associated with periodontal disease such as endometriosis.\cite{CDC2016} Despite these limitations, we believe that this study demonstrates the possible effect of IVF medications and super-physiological conditions on oral health. In addition, this work should increase awareness among infertility specialists and gynecologists to the possible adverse effects of sex steroid hormones on oral health.

**Acknowledgment**

The author would like to thank Dr. Aiman Zakarya MD, FRFCOG, FACOG for his help and support during the recruitment of patients attending his IVF clinic.

**Financial support and sponsorship**

Nil.

**Conflicts of interest**

There are no conflicts of interest.

**References**

1. CDC. Centre for Disease Control and Prevention (CDC) Report: (CDC’s National ART Surveillance System (NASS)−2016). CDC; 2016. Available from: http://www.cdc.gov/art/artdata/index.html.
2. Papageorgiou T, Guibert J, Goffinet F, Patrat C, Fulla Y, Janssens Y, et al. Percentile curves of serum estradiol levels during controlled ovarian stimulation in 905 cycles stimulated with recombinant FSH show that high estradiol is not detrimental to IVF outcome. Hum Reprod 2002;17:2846-50.
3. Sooriyamoorthy M, Gower DB. Hormonal influences on gingival tissue: Relationship to periodontal disease. J Clin Periodontol 1989;16:201-8.
4. Mariotti A. Sex steroid hormones and cell dynamics in the periodontium. Crit Rev Oral Biol Med 1994;5:27-53.
5. Machtei EE, Mahler D, Sanduri H, Peled M. The effect of menstrual cycle on periodontal health. J Periodontol 2004;75:408-12.
6. Miyazaki H, Yamashita Y, Shirahama R, Goto-Kimura K, Shimada N, Sogame A, et al. Periodontal condition of pregnant women assessed by CPITN. J Clin Periodontol 1991;18:751-4.
7. Hart R, Doherty DA, Pen nell CE, Newnham IA, Newnham JP. Periodontal disease: A potential modifiable risk factor limiting conception. Hum Reprod 2012;27:1332-42.
8. Klinger A, Hain B, Yaffe H, Schonberger O. Periodontal status of males attending an in vitro fertilization clinic. J Clin Periodontol 2011;38:542-6.
9. Haytaç MC, Cetin T, Seydaoglu G. The effects of ovulation induction during infertility treatment on gingival inflammation. J Periodontol 2004;75:805-10.
10. Pavlatou A, Tsami A, Vlahos N, Mantzavinos T, Vrotsos I. The effect of in vitro fertilization on gingival inflammation according to women’s periodontal status: Clinical data. J Int Acad
11. Lalasa G, Murthy KR, Pavankumar S, Raju GA. Periodontal status in infertile women attending in vitro fertilization clinics. Indian J Dent Res 2014;25:50-3.

12. Pavlatou A, Dokou P, Tsami A. Periodontal disease, infertility treatment and In Vitro Fertilization (IVF). J FIV Reprod Med Genet 2015;3:148.

13. Greene JC, Vermillion JR. The simplified oral hygiene index. J Am Dent Assoc 1964;68:7-13.

14. Löe H. The gingival index, the plaque index and the retention index systems. J Periodontol 1967;38:Suppl:610-6.

15. Mühlemann HR, Son S. Gingival sulcus bleeding – A leading symptom in initial gingivitis. Helv Odontol Acta 1971;15:107-13.

16. Clark DC, Chin Quee T, Bergeron MJ, Chan EC, Lautar-Lemay C, de Gruchy K, et al. Reliability of attachment level measurements using the cementoenamel junction and a plastic stent. J Periodontol 1987;58:115-8.

17. AlJehani YA. Risk factors of periodontal disease: Review of the literature. Int J Dent 2014;2014:182513.

18. Mealey BL, Moritz AJ. Hormonal influences: Effects of diabetes mellitus and endogenous female sex steroid hormones on the periodontium. Periodontol 2000 2003;32:59-81.

19. Mariotti AJ. Estrogen and extracellular matrix influence human gingival fibroblast proliferation and protein production. J Periodontol 2005;76:1391-7.

20. Nanba H, Nomura Y, Kinoshita M, Shimizu H, Ono K, Goto H, et al. Periodontal tissues and sex hormones. Effects of sex hormones on metabolism of fibroblasts derived from periodontal ligament. Nihon Shishubyo Gakkai Kaishi 1989;31:166-75.

21. Thomson ME, Pack AR. Effects of extended systemic and topical folate supplementation on gingivitis of pregnancy. J Clin Periodontol 1982;9:275-80.

22. Lindhe J, Bränemark PI, Lundskog J. Changes in vascular proliferation after local application of sex hormones. J Periodontal Res 1967;2:266-72.

23. ElAttar TM. Prostaglandin E2 in human gingiva in health and disease and its stimulation by female sex steroids. Prostaglandins 1976;11:331-41.

24. Ferris GM. Alteration in female sex hormones: Their effect on oral tissues and dental treatment. Compendium 1993;14:1558-64, 1566.

25. Chen TL, Aronow L, Feldman D. Glucocorticoid receptors and inhibition of bone cell growth in primary culture. Endocrinology 1977;100:619-28.

26. Güncü GN, Tözüm TF, Çağlayan F. Effects of endogenous sex hormones on the periodontium – Review of literature. Aust Dent J 2005;50:138-45.

27. Kavoussi SK, West BT, Taylor GW, Lebovic DI. Periodontal disease and endometriosis: Analysis of the National Health and Nutrition Examination Survey. Fertil Steril 2009;91:335-42.