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Do combined psychological stress examinations predict pregnancy outcome in an assisted reproductive technology program?

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Summary

Purpose of Investigation: To investigate prospectively if the pregnancy outcome in infertile women undergoing assisted reproductive technology (ART) is predictable by a combination of psychological stress examinations on the day of embryo/blastocyst transfer. *Materials and Methods*: From April 2012 to May 2012, 114 women aged 42 years old or less underwent transfer of morphologically-good embryo/blastocyst(s) in the present in vitro fertilization (IVF) center. Immediately before the transfer, salivary secretion was obtained and frozen. α-amylase and cortisol concentrations were quantified using biochemical assays. In addition, patients were asked to answer General Health Questionnaire 28 (GHQ28) and Zung's Self Rating Depression Scale (SDS) following transfer. The results were compared between the pregnant group and non-pregnant group. *Results*: There were no significant differences in the age of the infertile couples between the pregnant group and non-pregnant group as well as body mass index of the infertile women. The GHQ28 and SDS scores were similar between the two groups, as were the salivary α-amylase and cortisol concentrations. *Conclusion*: This prospective study failed to demonstrate the predictivity of the pregnancy outcome by psychological stress examinations in infertile women in an ART program, even though these tests were used in combination.

Key words: Assisted reproductive technology; Pregnancy outcome; Psychological tests; Salivary stress markers.

Introduction

The idea that psychological stress triggers major health problems, including cardiovascular and endocrinological diseases, has been widely accepted [1]. Studies also suggest the relationship between psychological stress and infertility including in vitro fertilization (IVF) failure and recurrent miscarriages [2, 3].

Psychological stress has been measured using questionnaires and endocrine markers such as α-amylase and cortisol. Recent advance in laboratory examinations demonstrated that the concentration of these molecules is accurately measurable in the saliva secretion as well as in the plasma [4, 5]. In the research of reproductive biology and pathology, these endocrine markers have been evaluated in the plasma samples, whereas a few studies utilized the secretion of the salivary glands.

Given the burden in infertility screening and treatment, repetition of venipuncture may further increase the psychological stress of the patients. Using less invasive salivary cortisol and α-amylase measurement and written questionnaires, we aimed to clarify the association between psychological stress and pregnancy outcome prospectively in an assisted reproductive technology (ART) program.

Materials and Methods

One-hundred and thirteen women who underwent embryo/blastocyst transfer in the present IVF center from April 2012 to May 2012 were enrolled in the study under informed consent [6]. The patients aged 43 years or more and/or with morphologically poor embryos/blastocysts were excluded from this study. This study was approved by our Institutional Review Board.

Immediately before the transfer, one ml of salivary secretion was obtained by their passive drooling. The sample was collected directly in a tube and stored at -20°C until measurement. α-amylase concentration was determined using a commercially available kinetic reaction assay [4], whereas cortisol concentration was quantified using a highly sensitive enzyme immunoassay [5]. The quantification was done in duplicate and the mean value was evaluated for comparison. During 30-minute bed rest following the transfer, patients were asked to answer two written psychological tests: General Health Questionnaire 28 (GHQ28) [7] and Zung's Self Rating Depression Scale (SDS) [8]. Serum HCG concentration was measured using an automated enzyme immunoassay on the 11th day following day-3 early cleavage embryo transfer or on the ninth day following day-5 blastocyst transfer. According to the manufacturer instruction, the values with two IU/L or more were regarded as a positive pregnancy test.

Statistical analysis was performed between the pregnant group and non-pregnant group. The scores and values were compared using Student's *t* test. A *p* value less than 0.05 was considered significantly different.

Results

There were no significant differences ($p > 0.13$) in the age of the infertile couples between the pregnant group (female partner 36.6 ± 3.4 years, and male partner 38.0 ± 5.0 years, mean ± SD) and non-pregnant group (female partner 37.4 ±

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Table 1. — *Characterization of the pregnant group and nonpregnant program in an ART program.*

	Pregnant group (n = 36)	Non-pregnant group (n = 77)	*p* value
Age (years)	36.6 ± 3.4	37.4 ± 4.3	0.32
Age, male partner (years)	38.0 ± 5.0	38.6 ± 4.7	0.51
Body mass index (kg/m^2)	22.0 ± 3.0	21.0 ± 3.0	0.13
GHQ28 score	5.0 ± 3.7	5.1 ± 4.9	0.90
SDS score	37.2 ± 6.3	36.7 ± 6.8	0.74
Salivary α-amylase concentrarion (μg/dl)	196.0 ± 144.6	202.0 ± 133.2	0.83
Salivary cortisol concentrarion (IU/ml)	0.16 ± 0.10	0.15 ± 0.02	0.46

All data are shown as mean ± standard deviation.

4.3 years, and male partner 38.6 ± 4.7 years), as well as body mass index of the infertile women (22.0 ± 3.0 kg/m^2 in the pregnant group vs 21.0 ± 3.0 kg/m^2 in the non-pregnant group (Table 1). The GHQ28 and SDS scores in the pregnant group (5.0 ± 3.7 and 37.2 ± 6.3, respectively) were comparable to that in the non-pregnant group (5.1 ± 4.9 and 36.7 ± 6.8, respectively) ($p > 0.74$). Finally, the salivary concentration of α-amylase (196.0 ± 144.6 mg/dl in the pregnant group *vs.* 202.0 ± 133.2 mg/dl in the non-pregnant group) and cortisol (5.1 ± 4.9 IU/ml in the pregnant group *vs.* 36.7 ± 6.8 IU/ml in the nonpregnant group, respectively) was also similar between the two groups ($p > 0.46$).

Discussion

The effect of psychological stress on reproduction remains controversial. While some studies support the negative impact of psychological stress on pregnancy, others deny it [9-15]. The discrepancy among the studies largely comes from the methodological variances and confounding factors. Researchers often use the time-to-pregnancy as a main outcome measure to assess the relationship between psychological factors and infertility [2, 9], but this parameter is frequently biased by diverse infertility etiologies such as ovarian reserve, tubal patency, intercourse frequency, and sperm count and motility. To reduce these biases, the authors limited the subjects to the infertile couples undergoing ART programs.

In this study, the authors did not find any significant differences in the molecular stress markers (salivary cortisol and/or α-amylase concentrations) and questionnaires (GHQ28 and SDS scores) between the pregnant and nonpregnant group following IVF-embryo transfer cycle. The strength of the present data is that prospective multiple measurements were adopted for evaluation. These results suggest that assessment of psychological stress in human reproduction is not easy and simplistic, although further investigations are required to reduce the intervention of the confounding factors. Some investigators reported that the level of these stress markers in infertile patients is higher than in fertile women [12, 15], implying that having infertility itself is a stressful condition.

Psychological stress measurement with four independent examinations including salivary secretory markers and written questionnaires failed to predict the pregnancy outcome in an ART program. These findings indicate that the results of IVF depend on various factors and larger studies are still required to detect the impact of psychological stress on pregnancy outcome.

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