Original Article

The Association of Anxiety and Perceived Stress with *In vitro* Fertilization Outcomes in Infertile Women: A Cross-sectional Study

Sara Abdoli[1,2], Amir Hossein Gholami[1], Seyedeh Zahra Masoumi[3], Roya Najafi-Vosough[4], Mozhdeh Azimi[5], Ensiyeh Jenabi[6], Sahar Aliabadi[5], Ali Reza Soltanian[7], Ali Ghaleiha[8], Shamim Pilehvari[9]

[1]Student Research Committee, Hamadan University of Medical Sciences, [2]Malayer School of Medical Science, Hamadan University of Medical Sciences, [3]Department of Midwifery and Reproductive Health, Mother and Child Care Research Center, School of Nursing and Midwifery, Hamadan University of Medical Sciences, [4]Department of Biostatistics and Epidemiology, School of Public Health, Hamadan University of Medical Sciences, [5]Hamadan University of Medical Sciences, [6]Mother and Child Care Research Center, Hamadan University of Medical Sciences, [7]Modeling of Noncommunicable Diseases Research Center, School of Public Health, Hamadan University of Medical Sciences, [8]Behavioral Disorders and Substance Abuse Research Center, Hamadan University of Medical Sciences, [9]Fertility and Infertility Research Center, Hamadan University of Medical Sciences, Hamadan, Iran

ABSTRACT

Background: The studies have shown that the effects of psychological distress on the outcome of *in vitro* fertilization (IVF) treatment are unclear, and there is still no conclusive empirical evidence that lower anxiety levels lead to better outcomes in assisted reproductive technique treatments. **Aim:** the present study was conducted to investigate the impact of perceived anxiety and stress on the outcome of IVF in infertile women in Western Iran. **Settings and Design:** The current study is a cross-sectional study. **Materials and Methods:** This study was conducted on infertile women who were candidates for IVF and visited the infertility center at Fatemiyeh Hospital in Hamadan City in the west of Iran from 8 July 2023, to 15 March 2024. The questionnaires were demographic and midwifery characteristics, the Spielberger State-Trait Anxiety Inventory and Perceived Stress Scale-14. **Statistical Analysis Used:** All statistical analyses were performed in SPSS software at a significance level of 0.05. **Results:** The stress was significantly associated with metaphase II eggs ($r = -0.487$), and the number of transferred embryos based on grade ($r = -0.562$), duration of marriage ($r = 0.628$), duration of infertility ($r = 0.595$) and duration of treatment ($r = 0.595$). There was no significant association between stress and endometrial thickness ($P = 0.189$). In addition, the anxiety was significantly associated with age ($r = 0.509$), husband›s age ($r = 0.436$), endometrial thickness ($r = -0.173$), metaphase II eggs ($r = -0.570$), and the number of transferred embryos based on grade ($r = -0.610$), duration of marriage ($r = 0.604$), duration of infertility ($r = 0.592$) and duration of treatment ($r = 0.604$). **Conclusion:** Mean stress and anxiety in infertile women were significantly lower in infertile women with successful IVF. Stress was significantly associated with age, spouse's age, metaphase stage oocytes, embryo grade, duration of marriage and infertility and duration of treatment.

KEYWORDS: *Anxiety, in vitro fertilization, Iran, stress*

Address for correspondence: Dr. Ensiyeh Jenabi,
Mother and Child Care Research Center, Hamadan University of Medical Sciences, Hamadan, Iran.
E-mail: en.jenabi@yahoo.com

INTRODUCTION

*I*nfertility is defined as the failure to achieve pregnancy after 1 year of regular unprotected intercourse.[1] Globally, approximately 15% of couples suffer from infertility.[2] Estimates of infertility vary across countries, ranging from 3.5% to 30%.[3] In Iran, the prevalence of infertility is between 10.3% and 24.9%.[4]

Assisted reproductive techniques (ART) are widely used in the treatment of infertility. *In vitro* fertilization (IVF) is the most common type of ART.[5] Women undergoing IVF treatment often experience depression and anxiety due to the uncertainty and low success rates of these treatments and endure higher levels of stress compared to the general population.[6] The psychological pressures and accompanying negative behavioural states, such as anxiety, can pose a threat to the outcomes of IVF treatment.[7]

Studies have shown that infertility is associated with stress, anxiety, life dissatisfaction and other psychological issues.[8,9] These problems can play a role in the outcomes of some infertility treatments.[10,11] In a study conducted by Aimagambetova *et al*. aimed at examining the impact of stress, depression and anxiety on IVF outcomes, the results indicated that levels of stress, anxiety and depression were higher in patients undergoing IVF treatment compared to the general population.[8]

Theoretically, stress can directly interfere with pregnancy through hormonal effects. Anxiety can exacerbate infertility by increasing cortisol and prolactin levels. Various mechanisms have been proposed for the impact of psychological pressures on fertility, including the disruption of gonadotropin secretion, local catecholamine effects on the uterus, impaired functioning of the fallopian tubes and disturbances in immunological processes involved in the maintenance and implantation of fertility.[12]

However, recent studies have shown that the effects of psychological distress on the outcome of IVF treatment are unclear, and there is still no conclusive empirical evidence that lower anxiety levels lead to better outcomes in ART treatments. Therefore, the present study was conducted to investigate the impact of perceived anxiety and stress on the outcome of IVF in infertile women in western Iran.

MATERIALS AND METHODS

Study design

The design of this study adhered to the STROBE reporting guidelines. The study protocol was approved by the Ethics Committee of Hamadan University of Medical Sciences (IR.UMSHA.REC.1402.015). Informed written consent was obtained from all participants.

Participants

The current study is a cross-sectional study conducted on infertile women who were candidates for IVF and visited the infertility infertility center at Fatemiyeh Hospital in Hamadan City from 8 July 2023, to 15 March 2024. This study was conducted based on a statement on adherence to Helsinki Declaration. Informed written consent was taken from all the subjects. To recruit participants, physicians identified women who were candidates for IVF and invited them to visit the infertility centre at their convenience, scheduling prior appointments.

Inclusion and exclusion criteria

The inclusion criteria for the study included the following: age range of 25–40 years, primary infertility diagnosed by a physician, women who have experienced the first cycle of IVF, not suffering from mental diseases diagnosed based on self-report, not suffering from diseases medical conditions such as diabetes, hypertension, thyroid, liver and heart diseases according to the file and self-report, no adverse event in the last month, no history of psychiatric hospitalisation, no addiction, no neurological or other progressive diseases and not been using neurological or psychiatric medications. The exclusion criteria included experiencing unfortunate events such as the death of loved ones during the study, achieving pregnancy naturally without ART methods during the treatment stages and developing physical or mental illness during the study.

Sample size

According to the results of Bapayeva *et al*.'s 2021 study,[13] the standard deviation (SD) of the stress score for IVF candidates was 18.4. For this study, it was assumed that the maximum significant difference would be approximately 3.39 ($d = 0.01 \times$ variance), with a Type I error rate of 0.05 and an estimated questionnaire dropout rate of about 20%. Consequently, the required sample size for the study was determined to be 136 participants.

Measurement tools

Demographic and midwifery characteristics

Demographic and midwifery characteristics were collected using a researcher-designed questionnaire. This questionnaire included information such as age, height, weight, occupation, age at marriage, length of marriage, level of education, level of income, health insurance coverage, duration and cause of infertility, endometrial thickness on the day of HCG injection, number of

dominant follicles on the day of HCG injection, number of metaphase II eggs, number of formed embryos, grading of transferred embryos and the result of IVF. To determine the validity of the midwifery information, the content validity method was employed. After reviewing various research articles in the field, the questionnaire was drafted and presented to 10 experts in gynaecology. Their feedback and suggestions were incorporated to refine the questionnaire, resulting in the final tool used for data collection.

Perceived Stress Scale-14

The Perceived Stress Scale (PSS) is a 14-item scale developed by Cohen *et al.* in 1983 to measure perceived stress over the past month.[14] It assesses how stressful an individual finds their life situations. The PSS uses a five-point Likert scale ranging from 0 (none) to 4 (very high), with items 5, 6, 7, 8, 9, 10 and 13 scored in reverse. The minimum and maximum possible scores on the scale are 0 and 56, respectively. A score above the cutoff point of 21.8 indicates high perceived stress. Cohen *et al.* reported a test–retest reliability of 0.85 for the scale. The scale's reliability, determined by calculating Cronbach's alpha for each subscale and the total score, ranged from 0.84 to 0.86. The validity and reliability of the PSS in Iran have been confirmed by Maroufizadeh *et al.*, who reported a Cronbach's alpha of 0.90.[15]

The Spielberger State-Trait Anxiety Inventory

The Spielberger State-Trait Anxiety Inventory (STAI) consists of 40 questions, with the first 20 measuring trait anxiety and the second 20 measuring state anxiety.[16] The trait anxiety scale includes 20 statements that evaluate an individual's feelings at 'this moment and time of response'. The state anxiety scale consists of 20 statements that measure people's general and ordinary feelings. For the trait anxiety responses, participants choose from the following options to indicate the intensity of their feelings: 1 (very low), 2 (low), 3 (high) and 4 (very high). To calculate the score for each of the two scales, the sum of the scores for the 20 statements on each scale is taken, with some statements scored in reverse. Consequently, the scores for both the trait and state anxiety scales range from 20 to 80. In Iran, Panahi Shahri confirmed the internal consistency of the STAI using Cronbach's alpha. For this sample, Cronbach's alpha was found to be 0.85 for the trait anxiety scale and 0.77 for the state anxiety scale.[17]

In vitro fertilization procedure

The ovulation stimulation method followed the standard procedure, utilising GnRH agonist in the luteal phase of the menstrual cycle and gonadotropin in the menstrual phase.[6] Daily injections of human menopausal gonadotropin and recombinant follicle-stimulating hormone were administered, with the daily dosage adjusted based on serial ultrasounds. The variables examined concerning patient prognosis included the number of predominant follicles > 16 mm on the day of HCG injection, endometrial thickness on the day of HCG injection, number of metaphase II eggs, number of formed embryos, grading of transferred embryos and pregnancy rate.

The psychological status of patients was evaluated at the start of the ovulation stimulation cycle through standard questionnaires, including demographic characteristics, the PSS-14 and the STAI. Participants were informed that they could contact the researcher in case of any issues or questions during the study. Following embryo transfer and pregnancy testing, participants reported the IVF results through telephone approximately 4–6 weeks after commencing the IVF process, and the questionnaire follow-up was subsequently completed.

Statistical analysis

The normality of variables was investigated using Kolmogorov–Smirnov test. The data were analysed using Spearman's, Mann–Whitney and Kruskal–Wallis tests. All statistical analyses were performed at a significance level of 0.05. The SPSS software (International Business Machines Corporation, New York State, USA) was used.

RESULTS

The mean (SD) age and husband age of infertile women were 34.35 (3.62) and 37.48 (3.93) years, respectively. The characteristics of the infertile women are given in Table 1. The majority of the infertile women had diplomas (34.6%) and were homemakers (88.2%). Embryo transfer was performed 4–6 weeks from the 1st day of injections for IVF. Grading was also performed using the Gardner system.

The number of transferred embryos based on grade in infertile women for Levels A, B and AB was 34 (25.0%), 35 (25.7%) and 67 (49.3%), respectively. The mean (SD) stress and anxiety of the infertile women were 41.06 (8.48) and 116.44 (22.23), respectively. Furthermore, the mean (SD) endometrial thickness, metaphase II eggs, number of embryos in the infertile women, duration of marriage, duration of infertility and duration of treatment were 9.12 (1.51), 8.24 (4.71), 6.26 (4.79), 2.06 (0.78), 2.18 (0.93) and 2.21 (1.01), respectively. Mean stress and anxiety were significantly higher amongst infertile homemakers compared to infertile employee women ($P < 0.001$) [Table 2]. In addition, mean stress and anxiety in infertile women were significantly lower in infertile women with successful IVF ($P < 0.001$).

Table 1: Demographic (qualitative) characteristics	
Variables	**n (%)**
Job	
Homemakers	120 (88.2)
Employee	16 (11.8)
Education	
Primary	24 (17.6)
High school	41 (30.1)
Diploma	47 (34.6)
university	24 (17.6)
Husband's job	
Employee	23 (16.9)
Unemployed	16 (11.7)
Self-employee	78 (57.4)
Manual worker	19 (14.0)
Education	
Primary	28 (20.6)
High school	33 (24.3)
Diploma	52 (38.2)
University	23 (16.9)

Table 2: Demographic Characteristics of Infertile Women (Quantitative Data)		
Variables	**Mean**	**SD**
Endometrial thickness	9.12	1.51
Metaphase ovum	8.24	4.71
Number of embryos	6.26	4.79
Duration of marriage	2.06	0.78
Duration of infertility	2.18	0.93
Duration of treatment	2.21	1.01

SD=Standard deviation

The mean stress and anxiety in infertile women were statistically significant between different levels of education and the husband's job ($P < 0.05$). In addition, the mean anxiety in infertile women was statistically significant between different levels of the husband's education ($P = 0.037$) [Table 3].

Relationships between stress and anxiety with demographic characteristics in infertile women are shown in Table 4. The stress was significantly associated with age ($r = 0.410$), husband's age ($r = 0.339$), metaphase II eggs ($r = −0.487$) and the number of transferred embryos based on grade ($r = −0.562$), duration of marriage ($r = 0.628$), duration of infertility ($r = 0.595$) and duration of treatment ($r = 0.595$). There was no significant association between stress and endometrial thickness ($P = 0.189$). In addition, the anxiety was significantly associated with age ($r = 0.509$), husband's age ($r = 0.436$), endometrial thickness ($r = −0.173$), metaphase II eggs ($r = −0.570$) and the number of transferred embryos based on grade ($r = −0.610$), duration of marriage ($r = 0.604$), duration of infertility ($r = 0.592$) and duration of treatment ($r = 0.604$).

DISCUSSION

A recent study examined the effect of perceived anxiety and stress on the outcome of IVF in infertile women. Stress was significantly associated with age, spouse's age, metaphase stage oocytes, embryo grade, duration of marriage and infertility and duration of treatment. However, there was no statistically significant relationship between stress and endometrial thickness. The results also indicated that fertility rates in patients were influenced by their levels of perceived anxiety and stress, with significantly lower mean levels of stress and anxiety observed in infertile women who had successful IVF outcomes.

Overall, mental health is a crucial factor for achieving pregnancy and having a healthy pregnancy period. Women undergoing IVF treatment are usually anxious and depressed due to infertility issues and the fear of treatment outcomes.[18] Anxiety causes the body to feel it is in a critical condition, which increases the production of the hormone cortisol. This hormone competes with testosterone, oestrogen and progesterone, which are the main reproductive hormones in both men and women.[19] Increased cortisol production leads to a decrease in fertility hormone levels. Couples who realise they have fertility problems face the reality that not only does infertility cause significant anxiety, but this anxiety itself can also impact their fertility.[20]

In recent years, the psychological aspects of infertility have garnered significant attention amongst healthcare providers and the general public.[9] Medical treatments can be a source of stress. Concerns about the long-term side effects of medications, the extensive time commitment for treatments and the high costs associated with infertility treatments are all factors that contribute to stress.[21] Some infertile couples may feel a sense of inadequacy and rejection, describing their infertility as the most distressing and stressful experience of their lives.[22]

According to the present study, the outcome of IVF is influenced by the factors of perceived anxiety and stress. However, the findings are inconsistent. Some clinical studies have examined the impact of psychological factors on IVF treatment. Aimagambetova *et al*. reported that higher levels of infertility-related stress are associated with lower IVF success rates.[8] In addition, Bapayeva *et al*. showed that the outcome of IVF is not significantly associated with depression and stress, whereas higher anxiety scale scores were negatively correlated with clinical pregnancy following IVF.[13] In a case–control study, the effect of pre-pregnancy depression, anxiety and stress on the likelihood of clinical pregnancy

Table 3: Correlation of mean stress and anxiety with demographic characteristics

Variables	Stress				Anxiety		
	Mean	SD	P		Mean	SD	P
Job							
Homemakers	42.61	7.75	<0.001		120.77	1.81	<0.001
Employee	29.43	2.68			6.82	1.70	
Education							
Primary	45.83	5.13	0.011		130.70	10.00	<0.001
High school	41.85	7.72			119.51	20.61	
Diploma	40.04	8.83			112.46	23.03	
university	36.95	9.54			104.75	24.41	
Husband's job							
Employee	36.91	9.62	0.022		106.78	25.84	0.001
Unemployed	45.50	3.77			131.75	6.98	
Self-employee	40.46	8.81			113.17	22.88	
Manual worker	44.84	5.02			128.68	8.83	
Husband's education							
Primary	42.60	6.06	0.095		123.42	17.01	0.037
High school	42.84	8.01			122.87	19.76	
Diploma	40.98	8.96			113.03	22.76	
University	36.82	9.46			106.43	25.52	
Embryos transferred based on grade							
A	40.17	9.08	0.897		113.88	22.12	0.759
B	41.28	7.83			117.62	23.33	
AB	41.40	8.57			117.13	21.93	
Successful IVF							
Negative	45.83	3.79	<0.001		129.51	7.67	<0.001
Positive	28.76	2.99			82.76	5.64	

SD=Standard deviation, IVF=In vitro fertilisation

Table 4: Bivariate correlations of stress and anxiety amongst infertile women

Variable	Variable			
	Stress		Anxiety	
	Correlation coefficient	P	Correlation coefficient	P
Age	0.410	<0.001*	0.509	<0.001*
Husband's age	0.339	<0.001*	0.436	<0.001*
Endometrial thickness	−0.113	0.189	−0.173	<0.001*
Metaphase ovum	−0.487	<0.001*	−0.570	<0.001*
Number of transferred embryos based on grade	−0.562	<0.001*	−0.610	<0.001*
Duration of marriage	0.628	<0.001*	0.604	<0.001*
Duration of infertility	0.595	<0.001*	0.592	<0.001*
Duration of treatment	0.595	<0.001*	0.604	<0.001*

*Significant at the 0.05 level

in infertile women undergoing IVF/ICSI treatment in a Chinese population was evaluated. The results indicated that pre-treatment psychological distress was not associated with pregnancy in infertile couples during their first treatment cycle.[23]

Furthermore, Liu *et al*. conducted a study aiming to examine the differences in anxiety and depression between Chinese infertile couples at different stages of IVF-embryo transfer (IVF-ET) and their relationship with IVF-ET outcomes. They found that anxiety and depression scores of infertile couples at different stages were not associated with IVF-ET outcomes.[11] Similarly, Maroufizadeh *et al*. reported in a prospective cohort study involving 142 women undergoing IVF treatment that there was no association between IVF outcomes and anxiety, depression or stress.[24] The differences between some studies and the present research may stem from variations in the studied population and the type of anxiety and stress assessment questionnaire used. In addition, cultural differences and the importance placed on having children can significantly impact individuals'

stress levels and, consequently, their response to treatment.

Consistent with the findings of the present study, Miller *et al.* demonstrated significant differences between the two groups in terms of pregnancy rates, indicating that anxiety and depression are associated with reduced likelihood of fertility in IVF cycles, and individuals without anxiety and depression have a higher chance of pregnancy following IVF.[25] Moreover, a systematic review also concluded that stress has a negative impact on IVF treatment. The ovarian retrieval stage is more affected by chronic and acute stress, whereas only chronic stress affects the embryo transfer stage. The stage of pregnancy rates had a weak association with stress.[26]

Limitations of the current study include self-reported data collection, which may introduce biases associated with this type of data, as well as the lack of comprehensive information on psychological issues and depression in women.

CONCLUSION

Mean stress and anxiety in infertile women were significantly lower in infertile women with successful IVF. Stress was significantly associated with age, spouse's age, metaphase stage oocytes, embryo grade, duration of marriage and infertility and duration of treatment. In Iran, laws supporting family and population youth are being pursued and implemented cohesively across various sectors. One secure and acceptable method to achieve this goal is by addressing infertility treatments to assist couples facing this challenge. Therefore, addressing women's mental health during this period is crucial to enhance policymaking and future planning, considering all aspects comprehensively. Moreover, it is essential to note that infertile women require psychological interventions and counselling. Providing appropriate counselling and necessary information about reproductive physiology, causes of infertility, familiarity with treatment processes and the side effects of medications can significantly reduce infertility-related anxiety.

Authors' contributions

EJ, AG, SM, MA, SA, SP, AG – Concept, design, manuscript preparation, data acquisition and analysis, editing and review; SA – Design and data acquisition; and AS and RN – literature search, manuscript preparation and editing.

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Conflicts of interest

There are no conflicts of interest.

Data availability statement

The data supporting the results can be made available on an Email request to en.jenabi@yahoo.com.

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