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Original Article

Predictors of Pregnancy Outcome for Libyan Infertile Women Underwent Intracytoplasmic Sperm Injection Cycle at Tripoli Infertility Center

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ABSTRACT

Intracytoplasmic Sperm Injection (ICSI) is a crucial treatment approach in assisted reproduction technology (ART) for infertility clinics. However, low pregnancy rates in ICSI cycles are a problem. Despite its potential, ICSI can be expensive, time-consuming, and cause concerns about gametes and newborn health. The study aims to identify factors predicting pregnancy rates among infertile couples undergoing Intracytoplasmic Sperm Injection cycle at Tripoli Infertility Center in Libya. This retrospective study reviewed all infertile couples' medical files at the Infertility Tripoli Centre from January to December 2023. A structured case sheet was used to collect data on female patients' age, causes of infertility, duration of infertility, previous pregnancy, antral follicles count, endometrial thickness, number of embryos transferred, and pregnancy outcomes. The study was divided into two groups based on pregnancy test results. Statistical analysis involved simple descriptive statistics, Chi square, and independent sample t-test. This study involved 90 women, with 35.6% having a positive pregnancy test. The pregnancy rate was 59.4% in women under 35, and decreased to 9.4% in those 40 or older. No significant differences were found in infertility duration or previous pregnancy (all $P > 0.05$). *Partner analysis revealed that 64.2% had asthenospermia, while endocrinopathies were reported by 41.8% of women. No significant differences were found in antral follicle count, endometrial thickness, oocyte number, or quality between pregnant and non-pregnant women* (all $P > 0.05$). Our findings suggest that *Maternal age has a significant impact on the success of ICSI, with advanced age in women negatively affecting ICSI outcomes.*

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INTRODUCTION

Infertility is one of the most significant complications in gynecology and is defined by the failure to achieve a clinical pregnancy after one year or more of regular unprotected sexual intercourse [1]. According to the WHO, infertility in the young population is the fifth-most serious global disability, affects about 8%-10% of couples worldwide, with 60-80 million couples experiencing infertility every year worldwide [2,3]. The estimated lifetime prevalence of infertility is lowest in the WHO Eastern Mediterranean Region (10.7%) and highest in the WHO Western Pacific Region (23.2%),

according to WHO data. The WHO African Region has the greatest estimated infertility prevalence (16.4%), while the Eastern Mediterranean Region has the lowest estimated infertility prevalence (10%) [4].

There are two types of infertility: primary and secondary. Women who have never become pregnant before are said to have primary infertility. There is at least one unsuccessful conception in secondary infertility [5]. Male infertility accounts for 20–30% of instances of infertility, female infertility accounts for 20–35%, combined male female factor infertility account for 25–40%, and unexplained causes is identified in 10–20% of cases [6]. The American Society of Reproductive Medicine (ASRM) standard infertility evaluation tests were: semen analysis, post-coital test, assessment of ovulation, hysterosalpingogram, and laparoscopy if indicated [7]. However, unexplained infertility (UI) stipulates that the initial evaluation performed on infertile couples should show that ovulation, appropriate sperm production, and normal fallopian tube patency have all been achieved [8]. Approximately 85% of infertile couples have an identifiable cause, and the remaining 15% of infertile couples have "unexplained infertility [9].

The consequences of involuntary childlessness are much more dramatic in developing countries and can create more wide-ranging societal problems compared to Western societies, particularly for women.[10]. Emotional losses can include denying parenthood, losing control of life, doubting one's womanhood, and losing friends [6].

Assisted reproductive technology includes all techniques involving the direct manipulation of oocytes and embryos outside the body. ART includes in vitro fertilization (IVF), intracytoplasmic sperm injection (ICSI), gamete intrafallopian transfer (GIFT), zygote intrafallopian transfer (ZIFT), cryopreserved embryo transfer, and the use of donor oocytes [11]. The international committee for assisted reproductive technologies monitoring was reported that ICSI was applied in 65% of cycles in Europe and this is highlighted in some areas of the world, ICSI is practiced in 100% of in vitro cycles [12]. Intracytoplasmic Sperm injection (ICSI) brings an operative technology in the fields of assisted reproduction technology (ART) [13]. Nowadays, this way becomes one of the most important and efficient treatment approaches which is used by infertility clinics.

One of problems in infertility centers is low rate of pregnancy in ICSI cycles. There is still an ongoing debate among reproductive embryologists and endocrinologists about using ICSI for the treatment of the infertile couples [13]. The emergence of assisted reproductive technology (ART) has been promising for infertile couples, but In Vitro Fertilization (IVF) success remains below 100% [14]. To increase the favorable outcome of ART, Intracytoplasmic Sperm Injection (ICSI) is used to treat severe male-factor infertility [15]. However, ICSI can be expensive, time-consuming, and cause unresolved concerns about gametes and newborn health [15] In addition, Infertility treatment by ICSI in Libya has not been adequately documented; this problem is crucial in countries like Libya. The current study was to determine the factors that predict the pregnancy rate among infertile couples undergo Intracytoplasmic Sperm Injection cycle (ICSI) at Tripoli Infertility Center in Tripoli, Libya.

METHODS

Study design and setting

This was a retrospective analytic study was conducting by reviewing all infertile couples' medical files were undergone ICSI trial at Infertility Tripoli Centre in Tripoli from 1st Jan to 31st Dec 2023.

Study tool

A structured case sheet was designed to collect the following data**;** female patients age, causes of infertility, duration of infertility, previous pregnancy (primary vs. secondary infertility). Antral follicles count (AFC), endometrial thickness, number of embryos transferred, and outcomes of pregnancy. Male partners were evaluated by seminal fluid analysis. Infertile women who were included in our study were subdivided into 2 groups according to the pregnancy test, which is used in the diagnostic evaluation. Pregnancy was defined by an increasing A beta human chorionic gonadotropin (B-hCG) at 15 days after oocyte retrieval. 1st group: those with a positive biochemical pregnancy test; 2nd group: those with a negative biochemical pregnancy test.

Statistical analysis

Simple descriptive statistics were used (mean ± standard deviation for quantitative variables and frequency with percentage for categorized variables). Categorical variables were compared using Chi square test and Fisher exact test. Continuous variables were compared using non-independent sample t test, and p-value < 0.05 was considered to be statistically significant.

Ethical considerations

Written consent approval was obtained from the research administration at the Tripoli Infertility Center to collect the data from patients' medical record, and confidentiality of the information was maintained throughout by excluding names in the study.

RESULTS

Demographic characteristics of pregnant women vs non pregnant women undergoing ICSI

Ninety women were recruited for this study. The pregnancy test was positive in (35.6%) of women**.** The general characteristics of all participants undergoing ICSI, divided into pregnant and nonpregnant groups, are provided in Table 1. The pregnancy rate was 59.4% in women under 35 years of age, while it decreased to 9.4% in women were 40 years or older (p=0.005). No significant statistical difference was seen in regards to duration of infertility, and previous pregnancy between women who got pregnant and those who didn't (P =0.618, P=0.169 respectively).

Table 1. Demographic characteristics of pregnant women vs non pregnant women undergoing ICSI.

Item	Pregnant (n=32)		Non -pregnant (n=58)		P -value
Age (year) (Mean± SD)	32.9 ± 5.71		38.2 ± 6.25		0.002[t]
Age group (year)	**F**	**(%)**	**F**	**(%)**	
Younger than 35	19	(59.4%)	18	(31.0%)	0.005[C]
35 -39	10	(31.3%)	17	(29.3%)	
≥ 40	3	(9.4%)	23	(39.7%)	
Duration of infertility (year) (Mean ±SD)	5.4 ± 2.92		5.9 ± 2.73		0.618[t]
Type of infertility	**F**	**(%)**	**F**	**(%)**	
Primary infertility	17	(53.1%)	40	(69.0%)	0.169[C]
Secondary infertility	15	(46.9%)	18	(31.0%)	

SD: Standard deviation. C: Chi square test. t: Independent sample T test. P < 0.05 is statistically significant.

Etiology of infertility of pregnant women vs non pregnant women undergoing ICSI

Sperm analysis of the partners of women showed that (64.2%) of them had asthenospermia, compared to (19.8%) with asthenteratozoospermia (4.8%) with oligospermia, (6.2%) with azoospermia, and just 3 (3.7%) with teratozoospermia. Furthermore, there was no apparent statistically significant difference between the two groups (p=0.084) (Table 2).

The most common female factors were endocrinopathies (hyperprolactinemia and thyroid disorders), which were reported by (41.8%) of the women, anovulation was reported by 25 (37.3%), tubal factors by 20 (29.9%), and endometriosis was reported by just six (9.0%) of the women, with no statistically significant difference between the two groups (p=0.278).

Table 2. Etiology of infertility of women undergoing ICSI

Item	Pregnant n=32		non-pregnant n=58		Total		
Cause of infertility	**F**	**(%)**	**F**	**(%)**	**F**	**(%)**	**P-value**
Male factor [M]							0.084[F]
Oligospermia	2	(16.7%)	10	(83.3%)	12	(14.8%)	
Asthenospermia	18	(34.6%)	34	(65.4%)	52	(64.2%)	
Teratozoospermia	0	(0.0%)	3	(100.0%)	3	(3.7%)	
Asthenteratozoospermia	7	(43.8%)	9	(56.3%)	16	(19.8%)	
Azoospermia	4	(80.0%)	1	(20.0%)	5	(6.2%)	
Female factor [M]							
Tubal	4	(20.0%)	16	(80.0%)	20	(29.9%)	0.278[F]
Endometriosis	3	(50.0%)	3	(50.0%)	6	(9.0%)	
Anovulation	11	(44.0%)	14	(56.0%)	25	(37.3%)	
Hormonal	11	(39.3%)	17	(60.7%)	28	(41.8%)	

M: Multiple responses were allowed. F: Fisher exact test.

Cycle parameters of pregnant women vs non pregnant women undergoing ICSI

Comparison of the mean antral follicles count and endometrial thickness in the pregnant with the non-pregnant women demonstrated in Table 3. There is no significant difference between the two groups (P = 0.238 and P = 0.270,

respectively). In addition, the means of oocytes number and quality were found to be no significant difference between two groups (P=0.186 and P=0.437, respectively).

Table 3. Cycle parameters of the of women undergoing ICSI

Variables	Pregnant	Non-Pregnant	P value
	Mean± SD	Mean± SD	
AFC	9.90 ± 4.92	8.52 ± 5.76	0.238[t]
Endometrial thickness (mm)	9.10 ± 1.56	8.66 ± 1.23	0270[t]
Number of good embryos	2.71 ± 1.18	2.26 ± 1.02	0.186[t]
Total number of embryos	2.31 ± 0.78	2.24 ± 1.42	0.437[t]

DISCUSSION

Assessment of pregnancy is the end-point that determines the success of infertility treatment. It is probably the most important outcome measure, and in the study of new treatment modalities in Libya, it is essential that the modalities being evaluated are effective in achieving a pregnancy and live birth in order to be able to counsel couples effectively about the prospect of treatment. To our knowledge, this is the first study in which the outcome for of pregnancy were measured in infertile women underwent the intracytoplasmic sperm program.

In this study, overall positive pregnancy rate was 35.6% which is comparable to previous studies investigation performed by Ashrafi *et al.*, in 2013 reporting pregnancy rate of 33.9% [13], and study in Pakistan in 2023 carried among 40 women aged between 20 – 40 underwent ICSI protocols a randomized clinical trial during two years from 2018 to 2020, reported that pregnancy rate of 30% [16], slightly higher than result obtained by Shulman *et al.*, study (23.9%) [17], and lower than those in Jordan by Khadra *et al.*, reported positive pregnancy rate was achieved in 44.3% [18].

Advanced women age has been associated with high rate of maternal and obstetrical complications such as maternal death, fetal and neonatal death, maternal hypertension, prematurity and operative delivery [13].

In our study, reported that poor ICSI outcomes have been associated with increasing of maternal age. Mukheef *et al.*, concluded that female ≤35 resulted in significantly better the intracytoplasmic sperm injection outcome.[19] In another study done by Zahir et al., the impact of maternal age on ICSI in Infertile couples was investigated, showing that pregnancy rate was significantly less in the older females [12]. Some centers of IVF restrict the maternal age for IVF as 43 years [10]. This explained that age-related infertility is primarily caused by diminished oocyte development competence and low ovarian reserve, leading to faulty fertilization, abortion, birth abnormalities, low implantation rates, reduced pregnancy rates, and increased loss rates in older women [12].

The outcomes of ICSI cycles for various infertility causes has yielded mixed results. Our results showed no significant effect of the etiology of infertility on the pregnancy rate, suggesting that ICSI is an effective option regardless of the cause. This is comparable to a previous study conducted in Iran, which included 1492 infertile women and aimed to evaluate the relationship between the ICSI outcome and the special cause of infertility. The results found no significant association between different causes of infertility and clinical outcomes [13].

The duration of infertility in our study has been found to have no a significant impact on the outcomes of women undergoing intracytoplasmic sperm injection (ICSI). An Egyptian study reported that did not find a significant difference in the duration of infertility between normal-weight and overweight/obese women [20]. Our findings suggest that the type of infertility is not a significant factor in the success of the ICSI trial. A range of studies have found that the type of infertility does not significantly impact the pregnancy rate in women undergoing ICSI. This result is consistent with a study conducted by Ashrafi et al., which found no association between different causes of infertility and clinical outcomes [13].

In the current study, it is notable that, no significant difference was found in endometrial thickness between pregnant and not-pregnant groups. This result is in agreement with the findings of an Iranian study which showed that no significant association between endometrial thickness and pregnancy outcome [21]. While, in a cross-sectional study by Hamdi et al. reported higher pregnancy rates in women with thicker endometrial linings, with also noting a higher pregnancy rate in those with a triple line pattern [22]. In Egyptian study found a correlation between endometrial sonographic parameters and ICSI success in normal responders [23].

In our study showed that the number of embryos and their quality do not significantly impact the success of intracytoplasmic sperm injection (ICSI) in women. A previous study was conducted in Malaysia conducted by Hafizi et al. found that increasing the number of collected oocytes and the cleavage rate could increase the chance of obtaining more high-grade embryos, but this did not necessarily lead to a higher success rate [24]. However, a study conducted Ajina et al. in Netherlands university medical centre in the Netherlands, revealed significantly lower a that the number

of 4-cell embryos transferred was a significant factor in predicting ICSI success, but the total number of embryos transferred did not have a significant influence [25].

Cryopreservation of embryos and oocytes is a common practice at our center. Infertile couples are required to sign documents recognizing their legal responsibilities regarding the cryopreservation of embryos or oocytes, retaining the right to make independent decisions. Specialized informed consent is crucial for fertility preservation procedures. It is important that couples are well-informed about their options and the associated risks. The use of cryopreservation is legally prohibited in our country if it exceeds two years from the date of freezing, or upon a patient's death, or the divorce of the couple. The study is constrained by its exclusive concentration on a single infertility center. The data provided is only pertinent to couples who have undergone multiple ART cycles and is restricted to fresh cycles. Additional research is necessary to improve the precision of these findings.

CONCLUSION

Maternal age significantly influences the success of ICSI, with older age in women adversely impacting the ICSI outcomes. No significant differences were found in pregnancy outcomes related to antral follicle count, endometrial thickness, or the number and quality of oocytes. It is essential that women at risk of premature ovarian failure due to gonadotoxic chemotherapy or radiation therapy be offered options for fertility preservation.

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Conflicts of Interest

There is no conflict of interest

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مؤشرات نتائج الحمل للنساء الليبيات المصابين بالعقم اللاتي خضعن لدورة حقن المجهري في مركز علاج العقم طرابلس

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المستخلص

الحقن المجهري هو نهج علاجي حاسم في تقنية المساعدة على الإنجاب. ومع ذلك ، فإن انخفاض معدلات الحمل في دورات الحقن المجهري يمثل مشكلة. على الرغم من إمكاناته ، يمكن أن يكون الحقن المجهري مكلفا ويستغرق وقتا طويلا ويسبب مخاوف بشأن الأمشاج وصحة الأطفال حديثي الولادة. الهدف من الدراسة هو تحديد العوامل التي تتنبأ بمعدلات الحمل بين الأزواج المصابين بالعقم الذين خضعوا لدورة الحقن المجهري في طرابلس . استعرضت هذه الدراسة بأثر رجعي جميع الملفات الطبية للأزواج المصابين بالعقم في مركز العقم في طرابلس من يناير إلى ديسمبر 2023. تم استخدام استبيان لجمع البيانات حول عمر المريضات ، وأسباب العقم ، ومدة العقم ، والحمل السابق ، وعدد الجريبات الغارية ، وسمك بطانة الرحم ، وعدد الأجنة المنقولة ، ونتائج الحمل. تم تقسيم الدراسة إلى مجموعتين بناء على نتائج اختبار الحمل. تضمن التحليل الإحصائي إحصاءات وصفية بسيطة ، ومربع كاي ، واختبار ت للعينة المستقلة.شملت هذه الدراسة 90 امرأة ، كان الحمل إيجابيا في 35.6٪منهم. كان معدل الحمل 59.4٪ لدى النساء دون سن 35 عاما ، وانخفض إلى 9.4٪ في سن 40 أو أكثر. لم يتم العثور على فروق ذات دلالة إحصائية في مدة العقم أو الحمل السابق (P > 0.05). كشف تحليل الزوج أن 64.2٪ يعانون من وهن النطاف ، بينما تم الإبلاغ عن اعتلالات الغدد الصماء من قبل 41.8٪ من النساء. لم يتم العثور على فروق ذات دلالة إحصائية في عدد الجريبات الغارية أو سمك بطانة الرحم أو عدد البويضات أو الجودة بين النساء الحوامل وغير الحوامل (جميعها P > 0.05). تشير النتائج التي توصلنا إليها إلى أن عمر الأم له تأثير كبير على نجاح الحقن المجهري ، حيث يؤثر تقدم العمر لدى النساء سلبا على نتائج الحقن المجهري.

الكلمات الدالة. الحقن المجهري، مركز طرابلس للعقم، المتنبئات، معدل الحمل.