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Effect of Administration of Single Dose GnRH Agonist in Luteal Phase on Implantation Rate of Fresh ICSI Cycles

Sally Alaa[1], Nadia Al-Hilli [2], Mufeda Ali Jwad[1]

[1]High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University, Baghdad, Iraq.
sallyalaa75@gmail.com
[2]Department of Obstetrics & Gynecology, College of Medicine, University of Babylon, Babylon, Iraq.

ABSTRACT

The luteal phase (LP) in the fresh ICSI cycle is insufficient, adequate LP support is one of the approved treatments for improving implantation and pregnancy rates. It is generally known that the LP is inadequate after ovarian stimulation due to negative from supra-physiological blood levels of steroids released by numerous corporal luteal, LH concentrations are low during the luteal phase. In this study, patients were divided into two groups: (40) patients as study group; those who received GnRHa (Decapeptil 0.1 mg), three days after embryo transfer, in addition to conventional luteal phase support (LPS) in the LP to increase the implantation and pregnancy rate in IVF; and their control group (40) received standard LPS only. On the second day of stimulation, blood samples for FSH, LH, TSH, E_2, and prolactin were taken. On the day of ovulation induction, measure E_2, progesterone, and LH; and on the day of embryo transfers, measure progesterone and LH. The overall characteristics of the patients in both groups were not significantly different. There was also no significant change in the number of total oocytes, mean of metaphase II oocytes percent, cleavage rate, grade I embryo percent, or serum hormones level between the study and control groups ($p > 0.05$). GnRH agonist treatment in the luteal phase improves clinical pregnancy and implantation rate in fresh ICSI cycles but is not statistically significant.

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KEYWORDS

Luteal Phase Support (LPS); GnRH; ICSI; FSH.

1. Introduction

Infertility is a big issue that many couples confront at some point in their lives. Reproductive medicine faces a difficult task in dealing with infertility issues. Infertility is estimated to affect 10% to 15% of the world's population (**Moridi et al., 2019** [1]). Infertility is defined as "the inability to achieve a clinical pregnancy following 12 months of regular, unprotected sexual intercourse" according to an updated worldwide dictionary on infertility and reproductive treatments (**Zegers-Hochschild et al., 2017** [2]). Fertility counseling, medical and/or surgical therapy of the underlying reason, fertility drugs, and assisted reproductive technologies (ARTs) are all part of infertility management (**Nardelli et al., 2014** [3]). All procedures that entail the in-vitro handling of both human oocytes and sperm or embryos for the aim of reproduction are referred to as assisted reproductive technologies (ART). Despite considerable advances in assisted reproductive technology (ART) that have overcome many of the underlying reasons for infertility, pregnancy rates remain low (**Zhang et al., 2013** [4]).

Positive yield in a series of IVF phases, including controlled ovarian stimulation (COS), ovum pick-up (OPU), fertilization, embryo transfer, and implantation, determines IVF success (**Tola, EN 2019** [5]). In ART therapy, successful implantation is a must-have result. Despite considerable efforts to improve, this result remains restricted. In reality, only around a third of instances involving one or more embryos result in a live delivery (**ESHRE, 2016** [6]), (**Chambers et al., 2017** [7]). Because the luteal phase in fresh cycles is deficient, adequate luteal phase support is one of the acceptable methods to increase implantation and pregnancy rates (**Humaidan et al., 2015** [8]), (**van der Linden et al., 2015** [9]). It is generally known that there is an inadequate luteal phase after ovarian stimulation (**Chau et al., 2019** [10]). Because of supra-physiological blood levels of steroids generated by many corpora lutea, LH concentrations are low during the luteal phase due to negative feedback on the pituitary gland (**Thomsen et al., 2018** [11]). LH is an important hormone for maintaining corpus luteum function and enhancing angiogenic factors,

growth factors, and cytokines that can help in implantation. LH inhibition causes early luteolysis or a considerable shortening of the luteal phase (**Chau et al., 2019** [10]). As aresult, fertility therapy with new cycles necessitates sufficient luteal support. GnRH agonist (GnRHa) has been shown to help with the luteal phase. On the one hand, it is thoughtthat GnRHa, given at the right dose, can maintain its stimulatory action, allowing LH production to continue through the luteal phase(**Fusi et al., 2019** [12]).

2. Materials and Methods

During the period November 2018 to September 2020, this prospective comparative study was conducted on 80 infertile women who were undergoing ICSI at Infertility Center of the High Institute for Infertility Diagnosis and Assisted Reproductive Technologies/ Reproductive Physiology, Al Nahrain University, Baghdad, Iraq. The Local Medical Ethical Committee of Al Nahrain University's High Institute for Infertility Diagnosis and Assisted Reproductive Technologies granted the current study ethical permission. In addition, each participant signed a formal informed consent form. In the ICSI laboratory of the High Institute for Infertility Diagnosis and Assisted Reproductive Technologies, the morphological examination of oocytes aspirated from the ovaries of infertile females and their resultant embryos was performed. The serum hormones were measured in a private laboratory using mini VIDAS. The participants in this study were 80 infertile females having ICSI cycles, ranging in age from 23 to 42 years old and infertility length from 4 to 13 years. In this study, women with both primary and secondary infertility were included. Some women have already had IVF/ICSI cycles, while others have not. A proportion of the women that were recruited had polycystic ovarian syndrome (PCOS). The fallopian tube was blocked in certain women. Following comprehensive standard assessment, a specific reason was not established in a proportion of couples, hence they were categorized as having unexplained infertility.

The study was designed to be a comparative prospective study. The selected 80 women are randomly divided into two groups:

A. **Study group:** In addition to regular luteal phase sport (LPS), 40 women underwent antagonist protocol with human chorionic gonadotrophin (HCG) trigger and got a single dose of GnRHa (Decapeptil 0.1 mg) on day 6 following ova pick-up (OPU).

B. **Control group:** 40 women undergo antagonist protocol with human chorionic gonadotrophin (HCG) trigger received standard luteal phase support only (LPS).

3. Results

The overall characteristics of the patients in both groups (the research group and the control group) were not significantly different, including age, weight, type of infertility (primary or secondary), reason of infertility, and length of infertility, as shown in **Table 1**. Follicle stimulating Hormone, Luteinizing Hormone, Estradiol, Thyroid stimulating Hormone; Prolactin and Progesterone levels were measured for both groups with 0.892 p value for the TSH being the highest response as shown in **Table 2**. Also, additional results were provided for the hormonal levels at the day of trigger of ET, with the highest p value for the Estradiol 0.793 at the day of trigger, these results were shown in **Table 3** and **Table *4.*** There was also no significant difference in total oocytes number, mean metaphase II oocytes percent, mean metaphase I percent, mean germinal vesicle oocyte percent, mean cleavage rate, mean grade I embryo percent, mean grade II embryos percent, or mean serum hormones level between the study and control groups ($p > 0.05$), as shown in **Table 5** and **Table 6**. Although no statistical evidence was found ($p > 0.05$), the study group that received GnRHa hormone 3 days after the embryo transfer, in addition to the standard luteal phase support (LPS) in the secretory phase of the fresh ICSI cycles, had a higher clinical pregnancy and implantation rate than the control group that received LPS only in the secretory phase. The implantation rate was 0.614 with the study group showing higher percentage of response than the control group, as shown in **Figure 1**. Pregnancy and twin pregnancy were compared between study groups as shown in **Table 7** and **Table 8**.

Table (1): Comparison of demographic features between study and control groups

Parameters	Control group	Study group	*p* value
Age (years) (Mean ± SD)	30.78±4.35	29.2±4.03	0.097 ₸
BMI (kg/m²) (Mean ± SD)	28.49±2.51	27.43±1.98	0.076 ₸
Duration of infertility (years) (Mean ± SD)	3.12±1.85	2.26±1.71	0.087 ₸
Type of infertility	Primary (20) Secondary (20)	Primary (20) Secondary (20)	1.00 ¥
Causes of infertility			
Male factor	13 (32.5%)	9 (22.5%)	0.484 ¥
Ovulatory	10 (25 %)	14 (35%)	
Tubal factor	10 (25%)	7 (17.5%)	
Unexplained	7 (17.5%)	10 (25%)	

SD: Standard deviation; BMI: Body mass Index; ₸: Independent sample t-test; ¥: Chi square.

Table (2): Comparison of basal hormonal levels between study and control groups

Hormonal level	Control group (Mean ± SD)	Study group (Mean ± SD)	*p* value
FSH (IU/L)	5.53 ± 1.41	5.35 ± 4.03	0.097 ₸
LH (IU/L)	4.09 ± 1.45	3.79 ± 1.7	0.547 ₸
Prolactin (IU/L)	13.86 ± 6.84	12.08±6.48	0.229 ₸
TSH (IU/L)	1.85 ± 0.76	1.88 ± 0.74	0.892 ₸
Progesterone (ng/ml)	0.49 ± 0.19	0.57 ± 0.27	0.156 ₸
E2 (pg/ ml)	39.89 ± 13.5	38.6 ± 9.18	0.616 ₸

SD: Standard deviation; BMI: body mass index: FSH: Follicle stimulating Hormone; LH: Luteinizing Hormone; E2: Estradiol; TSH: Thyroid stimulating Hormone; ₸: Independent sample t test.

Table (3): Comparison of hormonal levels at the day of trigger between study and control groups

Hormonal level	Control group (Mean ± SD)	Study group (Mean ± SD)	*p* value
E2 (pg/ mL)	1488 ± 334	1468 ± 327	0.793 Ŧ
PG (ng/ ml)	1.28 ± 0.45	1.1 ± 0.19	0.245 Ŧ
LH (IU/L)	1.89 ± 0.41	1.97 ± 0.36	0.386 Ŧ

LH: Luteinizing hormone at day of trigger; E2: Estradiol at day of trigger; PG: Progesterone at day of trigger; Ŧ: Independent sample t-test.

Table (4): Comparison of Progesterone and LH at day of ET between control and study groups

Hormonal level	Control group (Mean ± SD)	Study group (Mean ± SD)	*p* value
Progesterone at day of ET (ng/ ml)	16.57 ± 2.48	17.36 ± 3.69	0.313 Ŧ
LH at day of ET (IU/L)	16.41 ± 1.5	17.79 ± 0.98	0.322 Ŧ

Table (5): Comparison of oocytes characteristics between control and study group

Oocyte's characteristics	Control group (Mean ± SD)	Study group (Mean ± SD)	*p* value
Dominant oocytes	10.83 ± 2.86	11.56 ± 4.11	0.533 Ŧ
Metaphase II (MII)	7.91 ± 2.11	8.28 ± 2.03	0.424 Ŧ
Metaphase I (MI)	1.41 ± 1.56	1.66 ± 1.13	0.434 Ŧ
Germinal vesicle (GV)	1.16 ± 1.19	1.33 ± 0.99	0.182 Ŧ

Table (6): Comparison of embryo characteristics between study and control groups

Embryo characteristics	Control group (Mean ± SD)	Study group (Mean ± SD)	*p* value
Embryos number	2.8 ±1.07	3.08 ± 1.02	0.243 Ŧ
GI embryo no.	1.28 ± 0.45	1.38 ± 0.49	0.346 Ŧ
GII embryo no.	1.01 ± 0.98	1.46 ± 1.21	0.057 Ŧ

GI: Grade one; GII: Grade two; Ŧ: Independent sample t test.

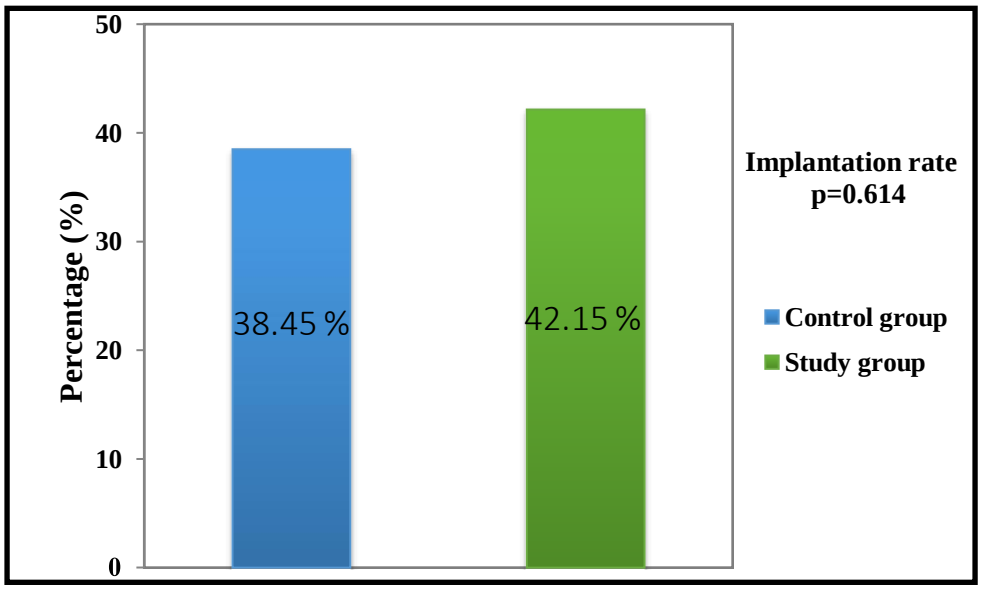

Figure (1): Comparison of implantation rate between study and control groups

Table (7): Comparison of pregnancy outcome between control and study group

Parameter	Control group (N.) (%)	Study group (N.) (%)	*p* value
Positive pregnancy	13 (32.5%)	17 (42.5 %)	0.356 ¥
Clinical pregnancy	5 (12.5 %)	12 (30 %)	0.056 ¥

Table (8): Comparison of twin pregnancy rate between control and study groups

Parameter	Control group (N=13)	Study group (N=17)	*p* value
Single pregnancy	12 (92.3%)	14 (82.4 %)	0.613 ¥
Twin pregnancy	1 (7.7 %)	3 (17.6 %)	
Total pregnancy	13 (100 %)	17(100 %)	

4. Discussion

Luteal phase insufficiency is a well-known problem in assisted reproductive technologies (ART). Many studies have suggested that GnRH agonists can be used as a routine luteal phase support (LPS). This study was designed to be a comparative prospective study that looked at the effect of a single dose of gonadotropin releasing hormone antagonist GnRHa (Decapeptil 0.1 mg) on the outcome of intracytoplasmic sperm injection in infertile women who were undergoing an antagonist protocol with HCG trigger on day 6 after ova pick-up or day 3 after embryo transfer in addition to standard luteal phase support. The fact that the participants were infertile women under the age of 42 with a regular menstrual cycle, both ovaries, and a normal uterus, as well as no history of chronic illness. In the current study, there was no statistically significant difference in demographic characteristics (age, BMI, duration, type, and causes of infertility) between the study and control groups, and all findings were comparable between the two groups to ensure statistical matching and reduce any variations that could affect the study's outcome. HCG level > 50 IU 14 days or more after embryo

transfer with or without visible gestational sac on ultrasound imaging indicates a positive pregnancy rate (**Benmachiche et al., 2017** [13]). Although the study group had a little higher positive pregnancy rate [17 (42.5%)] than the control group [13 (32.5%) in the current investigation. However, the finding was not statistically significant (P = 0.356). This finding was in line with prior research by(**Ata et al., 2008** [14]), (**Yildiz et al., 2014** [15]), (**Benmachiche et al., 2017** [13]). Clinical pregnancy was defined as a positive serum β-HCG test with ultrasound evidence of a gestational sac and fetal heart beat at 5 weeks after oocyte pick-up. In the current study, although clinical pregnancy rate in the study group [12 (30 %)] was higher than control group [5 (12.5 %)], but the difference was not significant (P = 0.056). Several studies have reached similar conclusions to ours. Multiple pregnancy rate was calculated as the percentage of pregnant women with a multifetal pregnancy per number of embryos transferred (**Han et al., 2015** [16]). Because of the expanding use of infertility therapy, particularly stimulation of ovulation with gonadotropins and in vitro fertilization, the incidence of high-order multiple gestations, defined as a pregnancy containing three or more infants, has increased (**Gleicher et al., 2000** [17]). Multiple pregnancy rates were equal in the GnRH agonist (single 0.1 mg triptorelin injection 6 days after ICSI) and placebo groups following ovarian stimulation with the extended GnRH agonist strategy, when transfer at least two embryos according to the Ata trial (**Ata et al., 2008** [14]). In Inamdar and Majumdar, there was no significant difference in the rate of multiple pregnancy in cycles induced with the extended GnRH agonist regimen between the GnRHa and placebo groups (**Majumdar and Inamdar, 2012** [18]). GnRHa may also have a direct influence on early embryos, enhancing implantation. Early meta-analyses suggested that mid-luteal GnRHa treatment had a favorable impact (**Oliveira et al., 2010** [19]), (**Kyrou et al., 2011** [20]). GnRHa may also have a direct effect on early embryos, helping them to implant. Early meta-analyses (**Oliveira et al., 2010** [19]) found that mid-luteal GnRHa therapy was helpful (**Kyrou et al., 2011** [20]).

6. Conclusions

Infertile women undergoing antagonist protocol, with hCG trigger had a higher clinical pregnancy and implantation rate of fresh ICSI cycles when given a mid-luteal single dose GnRH-agonist on day 6 after ova pickup or three days after embryo transfer(Day three embryo, grade one, one or more embryo transfer), in addition to standard luteal phase support, although it is statically non-significant.

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Author Contribution

Alaa, S performed the study, examined and reviewed results, and manuscript writing with the help and supervision of Jwad, MA, and Al-Hilli, N.

Conflict of Interest

The authors declare no conflict ofinterest.

Ethical Clearance

The study was approved by the Ethical Approval Committee.

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Authors at OrcID

Nadia Al-Hilli ***https://orcid.org/0000-0002-4486-9459***

Authors Biographies

Dr. Sally Alaa

She received the M.B.CH.B., D.O.G. from the College of medicine, University of Baghdad in 1998, 2008, repcetively. Her M.Sc. and Ph.D. in applied embroylogy from the High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University, Baghdad, Iraq.

Dr. Nadia Mudher Sulaiman Al-Hilli

She was born in Babylon, Iraq in 1976. She received the M.B.Ch.B. from College of Medicine, the University of Babylon in 1999, the Diploma in Obstetrics & Gynecology

in 2005, Iraqi Board in Obstetrics & Gynecology (FIBMS) in 2006, the Part I MRCOG in 2006, and the Certificate of minimal access surgery from the Iraqi Ministry of Health in 2017. Currently, she is an Assistant Professor and the Head of the department in Obstetrics & Gynecology department, College of Medicine, University of Babylon. She is also a lecturer and supervisor of Ph.D. students in the High Institute of Infertility and Assisted Reproductive Technologies since 2016. She is a member of the medical education committee at the University of Babylon, College of Medicine, and a member of the scientific committee of Obs. & Gyne. department at the University of Babylon, College of Medicine. She participated in more than, 25 Symposiums, 3 International Symposiums, 18 National Conferences, 15 International Conferences, 16 Workshops, and 9 Training Courses. She published more than 21 articles both local and international. She has more than 200 diagnostic & operative hysteroscopy operations & More than 200 diagnostic and operative laparoscopies performed for infertility patients.

Dr. Mufeda Ali Jwad

She received her MBChB. From the College of Medicine at the University of Baghdad in 1996. Her M.Sc. in Applied Embryology and her Ph.D. in Infertility and Clinical Reproduction were from the High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University in 2007 and 2018 respectively. She worked as a rotator in the Baghdad health department from 1996-1999. She worked in Gyn. & Obs. in Alsamawa general hospital and Babylon hospital from 2000-2003. She worked at the Babylon University, College of Medicine, anatomy and embryology department from 2003-2004. She has been working as a specialist physician and a consultant clinic at the High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al Nahrain University from 2008-2015. Currently, she is an assistant professor and specialist in infertility and clinical reproduction. She is the head of the clinical reproductive physiology department from 2019 till now. She has more than 25 published articles in national and international journals.