Progesterone versus combined estrogen and progesterone for luteal phase support (LPS) in women with unexplained infertility undergoing ICSI cycle: A randomized controlled, double-blinded study

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Abstract

Background : This study aims to determine the effectiveness and safety of adding estrogen and progesterone for luteal phase support to improve the live birth rate in women with unexplained infertility undergoing ICSI cycles using the long ovarian hyperstimulation protocol over the study period.

Methodology: This randomized, controlled, double-blinded study was conducted at the ART unit of Ain Shams University Maternity Hospital (ASUMH) from July 2020 till June 2021.It included 182 women, all of whom are suffering from unexplained infertility and underwent ICSI using the long protocol. Patients were randomly assigned into two groups: **Group A** ***(control): which*** consisted of patients who received vaginal progesterone supplementation (400mg twice a day), and **Group B** ***(study-estradiol group)*****:**2 mg of estradiol valerate were initiated orally along with progesterone, starting on the day of oocyte retrieval and continued until the end of first trimester. **Antenatal follow-up:** Patients with clinical conception did their antenatal care in the Ain Shams University Maternity Hospital outpatient clinic with follow-up of their outcome using a phone number.

Results: Regarding **main outcome measures,** statistical analysis of current results showed that the biochemical, clinical pregnancy, and live birth rates were all comparable in both groups. In group A, 34 (37.4%) had positive biochemical and clinical pregnancy compared with 40 (44.0%) patients in group B (p-value = 0.365). Twenty-six (28.6%) patients in Group A had live birth compared with 29 (31.9%) patients in Group B (p-value = 0.628). There was no statistically significant difference between both groups as regards the rate of twin pregnancy or CS delivery (p-value >0.999 and 0.628, respectively).

Conclusion: In women with unexplained infertility undergoing long protocol in assisted reproduction cycles, there were no significant differences between the relative effectiveness and safety of administering progesterone

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versus progesterone combined with estrogen for luteal phase support regarding biochemical, clinical pregnancy and live birth rates.

Key Words: luteal phase support, unexplained infertility, ICSI.

Background

The endometrium constitutes the inner layer of the uterus; it prevents adhesions between the opposed walls of the uterus, thereby maintaining its patency(1).The decidualized endometrium protects the embryo from maternal immunological rejection and provides nutrition before placental formation(2).

Even with high-quality embryos, the implantation rates in ART are still low, demonstrating the importance of impaired decidualization as a major cause of pregnancy failure (3).

In vitro fertilization (IVF) treatment usually involves ovarian stimulation (OS) with gonadotropins in addition to GnRH analogs to prevent premature luteinization and ovulation, as it is known that the use of GnRH analogs during OS may impair corpus luteum function, which results to suboptimal endometrial receptivity.Thus, using progesterone for LPS is an essential part of IVF treatment and is necessary to support implantation and increase pregnancy rates after fresh embryo transfer (4).

Still, there is a debate about what is best for LPS during IVF/ICSI cycle;. At the same time, some use progesterone-only protocols, and others prefer estrogen and progesterone protocols, which is still controversial.

Ceyhan et al.2008 (5) experienced higher pregnancy rates(56.5% vs. 61.9%) in their randomized study, which was performed on 60 patients, all of which are regular responders, and they demonstrated that in IVF cycles with antagonist protocol using estradiol in addition to progesterone led to better outcomes. Kwon et al.2013 (6) presented a randomized prospective study on 110 patients using antagonist protocol on IVF/ICSI cycles. They compared the use of progesterone-only versus estrogen and progesterone for LPS, and they demonstrated higher pregnancy rates with the combined approach (2.0% vs. 15.8%, p=0.035) and Also, this supplemental use significantly reduced the incidence of vaginal bleeding (7.4% vs. 27.8%, p=0.010).

Ismail Madkour et al. 2016 (7), in a recent prospective randomized study on 259 patients, showed no benefits from the additional use of estrogen to progesterone for luteal phase support in ICSI cycles. They demonstrated that ongoing pregnancy rates per embryo transfer conferred no significant difference in Group 1, with 32.7% and 32.7% in Group 2 (p=0.1). Also, there was no significant difference in implantation rates and abortion rates. In agreement with that, Pinheiro et al. 2017 (8) in his review comparing studies assessing the addition of estradiol to progesterone for LPS and its effects on pregnancy rates in IVF cycles using an antagonist protocol. They stated that only one study shows more successful embryo implantation in patients receiving estrogen progesterone combination. However, this success was not confirmed in any of the selected studies on pregnancy rate. Therefore, they emphasize the importance of further studies to clarify the role of estradiol in the luteal phase support in IVF cycles.

This study aims to determine the effectiveness and safety of adding estrogen and progesterone for luteal phase support to improve the live birth rate in women with unexplained infertility undergoing ICSI cycles using the long ovarian hyperstimulation protocol over the study period.

Patients and methods

This study was conducted at the ART unit of Ain Shams University Maternity Hospital

(ASUMH) from July 2020 to June 2021. This is a randomized, controlled (double-blinded) study. The Department of Obstetrics and Gynecology Council approved the study protocol and gained ethical approval from the Faculty of Medicine, Ain Shams University no (FWA 000017585).

The study was registered in the Pan-African Clinical Trial registry PACTR with ID 25717

Inclusion criteria are Couples with a diagnosis of unexplained infertility, planned treatment is the long protocol of ovarian stimulation in the context of ART, age group ranging between 18 years to 37 years, BMI ranging between 18.5 – 35, and every woman participating in the study signed an informed consent and had the right of withdrawal from the study at any time.

The following exclusion criteria were applied: previous uterine surgery, e.g., myomectomy, polypectomy, hydrosalpinx, uterine malformations, endometrial line < 8mm and/or not tri-laminar in sonography, endometriosis diagnosed by previous laparoscopy or ultrasound findings, previous preterm labor, recurrent pregnancy loss, HT, pre-eclampsia, eclampsia, unexplained IUFD, hypersensitivity to the used drugs, canceled cycles for poor or over response and failed fertilization, and withdrawal of the consent.

Primary outcome:

Live birth rate. number of births of neonates who showed any sign of life per 100 embryo transfer.

Secondary outcome:

Biochemical pregnancy: A pregnancy test was performed two weeks after the ICSI technique (Serum HCG).

Clinical pregnancy: The rate of clinical conception was confirmed with the presence of an intrauterine gestational sac with living embryo four weeks after embryo transfer.

Antenatal-care outcomes: side effects of estrogen as nausea, breast tenderness, headache, hypertension, venous thrombosis, predicted hyperstimulation syndrome during follow-up, spontaneous abortion rate, stillbirth, congenital anomalies, complication during pregnancy, e.g., hypertension, ectopic pregnancy, rupture membranes, DVT and delivery outcome were recorded.

Sample size justification:

The sample size was calculated using G*Power software version 3.1.2 for MS Windows, Franz Faul, Kiel University, Germany. Reviewing the literature, the available studies (10-12) addressed ongoing pregnancy rate as the primary outcome, and no studies addressed live birth rate as the primary outcome. Assuming a 5% difference between the two groups, a birth rate of 21% in group **A and 25%** in group **B**, a sample of 91 patients in each group would be enough to detect such a difference, if accurate, of 0.05 alpha errors & 0.80 power of the test.

Study interventions and procedures: All cases were subjected to detailed history taking, including age, parity, duration, and type of infertility, either primary or secondary, previous induction of ovulation, previous IVF, and previous ICSI in detail, i.e., when, how many times, age and outcomes, obstetric history, complications during pregnancy, and mode of delivery, general examination: weight, height, and body mass index (BMI).

Baseline evaluation:

The infertility workup was revised, including semen analysis, hormonal profile, hysterosalpingography, previous infertility treatment, and ART procedures.

Follicle-stimulating hormone (FSH), luteinizing hormone (LH), and estradiol (E2) were measured on day 2 of the cycle.

All participants underwent trans-vaginal sonography for endometrial lining assessment and antral follicle count (AFC).

Controlled ovarian hyperstimulation:

The long protocol was used for ovarian stimulation. In this protocol, the women were downregulated with a GnRH analog (Decapeptyl, Ferring, Egypt) administered 0.1 mg subcutaneously from day 21 of the previous menstrual cycle.

(E2) was analyzed on the second day of the menstrual cycle to assess pituitary suppression. If E2 was below 50 pg/ml, ovarian stimulation was done with human menopausal gonadotrophin hormone in the form of (Menogon®, Ferring, Egypt) 75IU vials from the second day of the menstrual cycle daily. The dose was calculated by the consultant in the IVF unit according to age, BMI, AFC, and response to previous IVF cycles.

Patients were followed by ultrasound folliculometry using trans-vaginal sonography (SONOACE X4) 7.5MHz on the 6th day of the menstrual cycle. Follow-up folliculometry was done according to follicle size with a further dose adjustment of the gonadotrophin dose according to the response calculated by the consultant in the IVF unit.

Triggering was done using HCG 10,000 units in the form of Choriomon®, IBSA, Egypt) when at least two follicles are exceeding 17mm in diameter.

Oocyte retrieval: The Oocytes were aspirated trans-vaginally under ultrasound guidance under general anesthesia 34 hours after triggering; fertilization was carried out by intra-cytoplasmic sperm injection (ICSI) technique, then embryo transfer of high-quality embryos guided by ultrasound 3 or 5 days after oocyte retrieval. The number of transferred embryos was determined by the IVF consultant; usually, two or three embryos were transferred or as available.

The Luteal support was started on the same day of oocyte retrieval. Patients were randomly assigned into two groups: **Group A** ***(control)***: consisted of patients who received vaginal progesterone supplementation (400mg twice a day) using (Prontogest®; Marcyrl, Egypt) combined with an identical placebo to the white tablets of Cycloprogynova tablets that were made at faculty of pharmacy Ain shams University starting on the day of oocyte retrieval and continued until the end of first trimester.

Group B ***(study-estradiol group)***:2 mg of estradiol valerate in the form of the white tablets of (CycloProgynova®; Bayer, Egypt) were initiated orally along with progesterone, starting on the day of oocyte retrieval and continued until the end of the first trimester.

Randomization was done by computer-generated random number sequence method into two groups, either group **(A)** or group **(B)**. We performed allocation concealment by sealed, opaque, sequentially numbered envelopes. This method was suitable for the current study.

Antenatal follow-up for patients with clinical conception did their antenatal care in Ain Shams University Maternity Hospital out-patient clinic with follow-up of their outcome using their phone numbers.

Blinding of patient and personnel:

For blinding, we used placebo preparation identical to the white tablets of Cycloprogynova made at the faculty of Pharmacy Ain Shams University prepared by the pharmacist in sealed opaque envelopes with serial numbers. The coding was kept with the pharmacist and revealed at the end of the study.

Statistical Analysis:

The collected data were revised, coded, tabulated, and introduced to a PC using the Statistical Package for Social Science

(SPSS 20.0.1 for Windows; Chicago, IL, 2001). Descriptive statistics for measured variables were expressed as a range, mean, and standard deviation (for metric data); range, median, inter-quarter range (for discrete data); number and proportion (for categorical data). The demographic data, primary and secondary outcomes of all patients were compared using a T-test (for quantitative parametric measures), Mann-Whitney's U-test (for quantitative non-parametric measures), Chi-square and Fisher exact test for categorical measures, sensitivity, specificity, positive predictive value, negative predictive value were calculated.

Results

Table 1 illustrates the demographic characteristics of both groups:

Table (1): Baseline evaluation among the studied cases

			Difference		95% CI		
Variable	Group A (n=91)	Group B (n=91)	Mean	SE	Lower	Upper	p-value†
Age (years), mean ± SD	30.3 ± 3.9	31.4 ± 4.4	-1.1	0.6	-2.3	0.1	0.076
BMI (kg/m^2), mean ± SD	28.6 ± 4.8	29.9 ± 3.9	-1.3	0.6	-2.6	0.0	0.043
Duration of infertility (years), mean ± SD	5.3 ± 3.2	5.2 ± 2.8	0.03	0.4	-0.8	0.9	0.941
Variable	Group A (n=91)		Group B (n=91)			P-value†	
Type of infertility, n (%)						0.048†	
Primary	62 (68.1%)		49 (53.8%)				
Secondary	29 (31.9%)		42 (46.2%)				
Parity, n (%)						0.003‡	
P0	83 (91.2%)		70 (76.9%)				
P1	8 (8.8%)		16 (17.6%)				
P2	0 (0.0%)		5 (5.5%)				
Abortions, n (%)						0.464†	
Nil	62 (68.1%)		52 (57.1%)				
1 Miscarriage	17 (18.7%)		26 (28.6%)				
2 Miscarriages	4 (4.4%)		7 (7.7%)				
≥3 Miscarriages	8 (8.8%)		6 (6.6%)				

†. Independent-samples t-test.

SD = standard deviation, SE = standard error, 95% CI = 95% confidence interval.

basal hormonal work up, ovarian reserve and semen analysis"). There was no statistically significant difference between both groups as regards baseline FSH (p-value = 0.557), LH (p-value = 0.346), TSH (p-value = 0.219), E2 (p-value = 0.650) or AMH (p-value = 0.234). Likewise, endometrial thickness, AFC, and the number of retrieved oocytes were comparable in both groups (p-value = 0.351, 0.537 and 0.300, respectively). Both groups were comparable regarding semen volume, sperm count and sperm motility (p-value = 0.195, 0.083 and 0.377, respectively). Mean abnormal forms was 90.4% (SD, 19.4%) versus 81.5% (SD, 27.8%) in Group A or Group B, respectively (p-value = 0.013).

Controlled ovarian stimulation data of the study participants: Table 2 illustrates induction characteristics, ovulation characteristics, fertilization characteristics, and embryo transfer characteristics among studied cases.

Induction Characteristics	Group A	Group B	P-value
Long Protocol	Group A (n=91)	Group B (n=91)	
Stimulation duration (days) **Mean±SD** **Range**	13.2±2.8 8.0–21.0	13.2±2.8 8.0–21.0	<0.999
Total dose (IU) **Mean±SD** **Range**	3392.6±1322.3 750.0–7200	3392.6±1322.3 750.0–7200	<0.999
AFC **Mean±SD** **Range**	13.7±6.3 4.0–39.0	13.7±6.3 4.0–39.0	<0.999
Ovulation characteristics:			
Variable	**Group A**	**Group B**	**P-value**
Ovulation	N 86	N 84	
Day of ovum pickup Mean±SD **Range**	15.2±2.9 10.0–23.0	15.1±2.6 10.0–22.0	0.911
Number of expected ovum pickup **Mean±SD** **Range**	10.1±6.5 2.0–31.0	10.0±6.3 2.0–30.0	0.922
Number of oocytes retrievedMean±SD **Range**	9.3±6.1 1.0–27.0	9.2±6.0 1.0–26.0	0.981
Number of M2 oocytes **Mean±SD** **Range**	7.5±5.1 1.0–24.0	7.5±5.0 1.0–23.0	0.999
Fertilization characteristics:			
Variable	**Group A**	**Group B**	**P-value**
Fertilization	83	84	
Number of fertilized oocytes Mean±SD range	5.0±3.4 1.0–19.0	5.1±3.6 1.0–20.0	0.982
Fertilization rate **Mean±SD** **Range**	71.4±23.0 14.1–99.0	71.6±23.1 14.3–100.0	0.971
Embryo transfer characteristics:			
Variable	**Group A**	**Group B**	**P-value**
Embryo transfer	82	81	
Day of embryo transfer **Mean±SD** **Range**	3.9±1.0 3.0–5.0	3.9±1.0 3.0–5.0	<0.999
Number of transferred embryos **Mean±SD** **Range**	2.2±0.7 1.0–4.0	2.2±0.7 1.0–4.0	<0.999

†. Independent-samples t-test.
SD = standard deviation, SE = standard error, 95% CI = 95% confidence interval.

Table 3: illustrated main outcomes , maternal complications and fetal complications:

Variable	Group A (n=91)	Group B (n=91)	P-value†
Biochemical pregnancy, n (%)	34 (37.4%)	40 (44.0%)	0.365
Clinical pregnancy, n (%)	34 (37.4%)	40 (44.0%)	0.365
Twin, n (%)	6 (6.6%)	6 (6.6%)	>0.999
Live birth, n (%)	26 (28.6%)	29 (31.9%)	0.628
CS delivery, n (%)	26 (28.6%)	29 (31.9%)	0.628
Incidence of maternal adverse outcomes in both groups:			
Nausea, n (%)	31 (34.1%)	34 (37.4%)	0.643†
Vomiting, n (%)	24 (26.4%)	22 (24.2%)	0.733†
Headache, n (%)	13 (14.3%)	24 (26.4%)	0.043†
Gestational DM, n (%)	4 (4.4%)	5 (5.5%)	>0.999‡
Gestational hypertension, n (%)	8 (8.8%)	10 (11.0%)	0.619†
Withdrawn because of adverse effects, n (%)	7 (7.7%)	4 (4.4%)	0.351†
DVT, n (%)	0 (0.0%)	0 (0.0%)	NC
APH, n (%)	0 (0.0%)	0 (0.0%)	NC
Complications	**Group A**	**Group B**	**p-value**
Ovarian hyperstimulation syndrome	8	8	<0.999
Hetero-tropic pregnancy	0	0.0	NC
Multiple pregnancy	6	6	<0.999
Mortality	0	0.0	NC
Fetal complications among the studied cases			
Early fetal loss (miscarriage), n (%)	8 (8.8%)	11 (12.1%)	0.467†
PTD, n (%)	2 (2.2%)	6 (6.6%)	0.278‡
IUGR, n (%)	1 (1.1%)	2 (2.2%)	>0.999‡
Macrosomia, n (%)	0 (0.0%)	1 (1.1%)	>0.999‡
PROM, n (%)	2 (2.2%)	4 (4.4%)	0.682‡

Chi-squared test for trend.

Discussion

We found that in women with unexplained infertility undergoing long protocol in assisted reproduction cycles, there were no significant differences between the relative effectiveness and safety of administering progesterone versus progesterone combined with estrogen for luteal phase support regarding biochemical, clinical pregnancy, and live birth rates.

Regarding main outcome measures, statistical analysis of current results showed that the biochemical, clinical pregnancy, and live birth rates were all comparable in both groups. In group A, 34 (37.4%) had positive biochemical and clinical pregnancy compared with 40 (44.0%) patients in group B (p-value = 0.365). Twenty-six (28.6%) patients in Group A had live birth compared with 29 (31.9%) patients in Group B (p-value = 0.628). There was no statistically significant difference between both groups as regards the rate of twin pregnancy or CS delivery (p-value >0.999 and 0.628, respectively).

Regarding the incidence of maternal adverse outcomes, current results showed that significantly more patients in group B

complained of headaches (24 [26.4%] versus 13 [14.3%], p-value = 0.043). Otherwise, there was no statistically significant difference between both groups as regards the incidence of nausea (p-value = 0.643), vomiting (p-value = 0.733), gestational DM (p-value >0.999) or gestational hypertension (p-value = 0.619). None of the patients in either group had DVT or APH. The rate of withdrawal from the study because of medication-related adverse effects was comparable in both groups (7 [7.7%] versus 4 [4.4%] in Group A or Group B, respectively, p-value = 0.043).

Regarding the incidence of fetal adverse outcomes, statistical analysis of current results showed that there was no statistically significant difference between both groups as regards the incidence of early fetal loss (p-value = 0.467), PTD (p-value = 0.278), IUGR (p-value >0.999), macrosomia (p-value >0.999) or PROM (p-value = 0.682). None of the patients in either group had fetal anomalies.

Comparison of our results to similar studies

Çakar and his colleagues conducted a case-control study to evaluate the effect of combined use of oral estrogen (E2) and vaginal progesterone (P) for LPS in antagonist (ICSI) cycles. A total of 176 patients were enrolled. Once a day, progesterone 90mg vaginal gel and micronized E2 of 4 mg/day was started from the day of oocyte pick up and continued to the 12th day of embryo transfer. Group 1 (n=79) patients received E2 +P for luteal phase support. In group 2(n=97) patients, only P 90mg vaginal gel was used. They agreed with our study and stated that no significant difference existed between group 1 and group 2 in means of pregnancy rate (26.58% n = 21 vs. 24.74%n = 24) (p = .781), clinical pregnancy rate (26.58% n = 21 vs. 20.62%n = 20) (p = .352) and implantation rate (22.8% n = 21 vs. 16.9% n = 20) (p = .298), the incidence of luteal vaginal bleeding (8.86% n = 7 vs. 8.25% n = 8) (p =.885). They also agreed with our study and stated that no significant difference existed between group 1 and group 2 in means of early pregnancy loss rate (6.33% n = 5 vs. 6.19% n = 6) (p = .969) (9).

Madkour and his colleagues conducted a randomized controlled study to compare pregnancy outcomes in 220 patients undergoing antagonist (ICSI) cycles protocol. The patients were randomly assigned into two equal groups to receive either vaginal progesterone alone (90mg once daily) starting on the day of oocyte retrieval for up to 12 weeks if pregnancy occurred or estradiol addition (2mg twice daily) starting on the same day and continuing up to seven weeks (fetal viability scan). They agreed with the current study and stated that early pregnancy loss rates were comparable with 6.3% and 7.2% for groups 1 and 2, respectively (p value=0.4). They also agreed with the current study and stated that pregnancy rate per embryo transfer did not differ between group 1 (progesterone) (39.09%) compared to group 2 (progesterone/ E2 group) (43.63%) (p value=0.3). Similarly, both groups gave comparable ongoing pregnancy rates per embryo transfer with32.7% in group 1 compared to 36.3% in group 2(p value=0.1) (7)

Lin and his colleagues conducted a prospective randomized controlled study on 402 patients to explore whether oral oestradiol (E2) supplementation (6 mg) in the luteal phase is beneficial to the outcome of patients undergoing gonadotrophin-releasing hormone agonist (GnRHa)long protocol in vitro fertilization (IVF)/(ICSI) cycles. In total, 402 patients were prospectively randomized to receive either progesterone injection plus oral E2 supplementation (Group A, n = 202) or progesterone injection alone (Group B, n = 200) for LPS after oocyte retrieval. They agreed with our study and stated that the cycle outcomes, including clinical pregnancy rate, implantation rate, miscarriage rate, and moderate OHSS rate, were comparable between the groups. (10)

Engmannand his colleagues agreed with

current study and stated there were no significant differences in the implantation (56/210 [26.7%] vs. 64/203 [31.5%]), clinical pregnancy (42/84 [50%] vs. 52/82 [63.4%]), and ongoing pregnancy rates (40/84 [47.6%] vs. 46/82 [56.1%]) between the study and control groups, respectively. One hundred sixty-six patients undergoing their first IVF treatment cycle were enrolled in a prospective randomized controlled trial. Patients underwent three different protocols for controlled ovarian hyperstimulation for IVF treatment with long GnRH agonist suppression, use of GnRH antagonist, or a microdose GnRH agonist protocol. LPS was in the form of IM P. Patients randomized into the study group (n = 84) received E2 supplementation in the form of vaginal estrace 2 mg twice a day starting on the day of ET. Patients randomized to the control group (n = 82) received no E2 supplementation. (11)

Serna and his colleagues agreed with current study and stated that there were no statistically significant differences in terms of implantation rate (34.9% [51 of 146] vs. 28.9% [41 of 142]), ongoing pregnancy rate 42% ([34 of 81]vs. 41.8% [33 of 79]), early pregnancy loss (15% [6 of 40] vs. 13.2% [5 of 38]), or multiple pregnancy rate(28.6% [12 of 42] vs. 24.4% [10/41]) in patients receiving P versus E2 + P.(12)

Against the current study, Drakakis and his colleagues stated that estradiol supplementation during the luteal phase in women undergoing IVF/ICSI-ET benefits the outcome without adverse effects. In this prospective, randomized study, they studied patients undergoing IVF/ICSI with controlled ovarian hyperstimulation using a gonadotropin-releasing hormone agonist/ human recombinant gonadotropin long protocol. A total of 77 patients were included in the study. The first group received estrogen and progesterone supplementation from the day of oocyte retrieval (N=39), and the second group(N=38) took only progesterone supplementation during the luteal phase. From the 24 cases with successful outcomes, 75% were from Group 1 that received estradiol supplementation, and 25% were from Group 2 with no estradiol ($p<0.05$). but there was a tendency towards a higher abortion rate in Group 1 (10.3% vs.2.6%). The implantation and pregnancy rates were significantly increased in the group with estradiol supplementation (implantation rate: 10.2vs. 4.0%, pregnancy rate: 46.1 vs. 15.8%; $p<0.05$ for both) (13). These differences can be attributed to the small sample size included in Drakakis's study.

Strengths and limitations of our study

Our strength point is that all clinical assessment and assessment of study outcomes were done by the same team. Blinding of patients and personnel was achieved. The limitation of our study is the relatively small number of patients and it is a single and not multicenter study.

The clinical implication of this study is that we did not find evidence of the benefit of adding estrogen to progesterone for luteal phase support in women undergoing ICSI using the long protocol.

Recommendation for future research

We recommend further future studies with larger sample sizes to demonstrate what is best for luteal phase support in ICSI cycles with different ovulation induction protocols.

Conclusion

In women with unexplained infertility undergoing long protocol in assisted reproduction cycles, there were no significant differences between the relative effectiveness and safety of administering progesterone versus progesterone combined with estrogen for luteal phase support regarding biochemical, clinical pregnancy and live birth rates.

List of abbreviations

1. LPS : luteal phase support .
2. ICSI : intracytoplasmic sperm injection .
3. ART: assisted reproductive techniques .
4. ASUMH : Ain Shams university maternity hospital .
5. IVF: in vitro fertilization .
6. GnRH: gonadotrophin releasing hormone
7. BMI: body mass index .
8. FSH: follicle stimulating hormone.
9. LH: luteinizing hormone .
10. E2 : estradiol.
11. AFC: antral follicle count.
12. HCG: human chorionic gonadotrophin.
13. SD: standard deviation .
14. IU : international unit .
15. ET: embryo transfer .

Declarations

Ethical: The Obstetrics and Gynecology Department Council approved the study protocol and ethically was approved by the Ethical Research Committee (Faculty of Medicine, Ain Shams University). FMASU 000017585

Availability of data: data will be available from the corresponding author upon reasonable request.

Conflict of interest: The authors declare that they have no competing interests.

Funding: not applicable.

Acknowledgments: not applicable.

Authors contributions

MH did the literature review, was participated in data collection and follow-up of cases

SS participated in the literature reviewand revision of the manuscript.

YA was responsible for data collection, statistics, and gathering scientific data.

WK was responsible for revising all data and writing the manuscript.

References

1. Tinelli A, Mynbaev OA, Vergara D, Di Tommaso S, Gerli S, Favilli A, et al. Uterine-preserving operative therapy of uterus myomatosus. Hysterectomy: A Comprehensive Surgical Approach. 2018:429-66.
2. Mori M, Bogdan A, Balassa T, Csabai T, Szekeres-Bartho J. The decidua maternal bed embracing the embryo-maintains the pregnancy. SeminImmunopathol. 2016;38:635-649.
3. Coulam C. What about superfertility, decidualization, and natural selection?. J Assist Reprod Genet. 2016;33:577-80.
4. Delcour C, Robin G, Delesalle AS, Drumez E, Plouvier P, Dewailly D, et al. Weekly intramuscular progesterone for luteal phase support in women receiving oocyte donation is associated with a decreased miscarriage rate. Reproduct Biomed Online. 2019; 39:446–451.
5. Ceyhan ST, BasaranM, Kemal Duru N, Yilmaz A, GoktolgaU, Baser I. Use of luteal estrogen supplementation in normal responder patients treated with fixed multi-doseGnRH antagonist: a prospective randomized controlled-study. Fertility and Sterility 2008; 89(6):1827–30.
6. Kwon SK, Kim CH, Lee KH, Jeon IK, Ahn JW, Kim SH, et al. Luteal estradiol supplementation in gonadotropin-releasing hormone antagonist cycles for infertile patients in vitro fertilization. ClinExpReprod Med. 2013;40:131–134.
7. Madkour WA, Noah B, Abdel HamidAM, Zaheer H, Al-Bahr A, Shaeer, M, et al. Luteal phase support with estradiol and progesterone versus progesterone

alone in GnRH antagonist ICSI cycles: a randomized controlled study. Human Fertility. 2016; 19(2), 142-49.

8. Pinheiro L, Cândido P, Moreto TC, Almeida W, & Castro EC . Estradiol use in the luteal phase and its effects on pregnancy rates in IVF cycles with GnRH antagonist: a systematic review. JBRA assisted reproduction.2017; 21(3):247–250.
9. ÇakarE, Tasan HA, Kumru P, Cogendez E, Usal NT, Kutlu HT et al. Combined use of oestradiol and progesterone to support luteal phase in antagonist intracytoplasmic sperm injection cycles of normo-responder women: a case-control study. Journal of Obstetrics and Gynaecology. 2020; 40(2): 264-69.
10. LinH, LiY, Li L, Wang W, Zhang Q, Chen X et al. Oral oestradiol supplementation as luteal support in IVF/ICSI cycles: a prospective, randomized controlled study. European Journal of Obstetrics & Gynecology and Reproductive Biology,2013;167(2): 171175.
11. Engmann, L, DiLuig A, Schmid, D, Benadiva C, Maier D, Nulsen J. The effect of luteal phase vaginal estradiol supplementation on the success of in vitro fertilization treatment: a prospective randomized study. Fertility and sterility.2008; 89(3), 554-561.
12. Serna J, CholquevilqueJL, Cela V, Martínez-Salazar J, RequenaA, Garcia-Velasco J A . Estradiol supplementation during the luteal phase of IVF-ICSI patients: a randomized, controlled trial. Fertility and sterility. 2008; 90(6), 2190-2195.
13. Drakakis P, Loutradis D, Vomvolaki E, Stefanidis K, Kiapekou E, Anagnostou Eet al.. Luteal estrogen supplementation in stimulated cycles may improve the pregnancy rate in patients undergoing in vitro fertilization/intracytoplasmic sperm injection–embryo transfer. Gynecological Endocrinology. 2007;, 23(11), 645-652.