

# Effect of Oral Estradiol Valerate versus Vaginal Sildenafil on Endometrial Receptivity Evaluated by Ultrasound and Pregnancy Rate in Iraqi Infertile Females

Sundus Ali Dawood<sup>1</sup>, Dr. Huda Ali Hussaini<sup>2</sup> Dr. Mufeda Ali<sup>1</sup>.

<sup>1</sup>High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al-Nahrain University, Baghdad, IRAQ.

<sup>2</sup>Collage of medicine, Al-Nahrain University, Baghdad, IRAQ.

**Corresponding author:** Sundus Ali Dawood

E-mail: [raadreshan@yahoo.com](mailto:raadreshan@yahoo.com)

Article History:

Submitted: 15.04.2020

Revised: 13.05.2020

Accepted: 25.06.2020

## ABSTRACT

Female causes account for 30 percent of the overall infertility cases, five percent were attributed to uterine factors. Endometrial thickness (ET) is one of the best predictors of implantation rate and continued success rate for pregnancy. There were possible beneficial effects of sildenafil citrate or estradiol on endometrial receptivity so some reports discussed these potential effects

**The aim:** of this study is to compare the effect of estradiol valerate and vaginal sildenafil citrate on endometrial receptivity (endometrial thickness, serum Vascular Endothelial Growth Factor) and pregnancy rate in infertile Iraqi women

**Subjects and Methods:** Sixty infertile women were included in this comparative prospective study undergoing stimulated cycles and timed intercourse. Patients diagnosed previously to have thin endometrium were divided randomly into two groups. In group A, thirty patients were given estradiol valerate 2 mg tablet 12 hourly. In group B, 30 patients were given sildenafil citrate 25 mg vaginally every 6 hours. Patients were evaluated by using trans-vaginal sonography (TVS) to determine and compare the effects of estradiol valerate and vaginal sildenafil citrate on endometrial thickness, pattern, and serum assessment of Vascular Endothelial Growth Factor VEGF levels and pregnancy outcome in both groups.

**Results:** Mean endometrial thickness at HCG day was 9.19 mm in

estradiol treated group and 8.86 mm in sildenafil treated group which was more in estradiol group but statistically insignificant ( $p > 0.05$ ). And endometrial triple line pattern in the Estradiol and sildenafil citrate groups was 96.6%, and 90% respectively. VEGF level in estradiol treated group was (220.5 ng/l) compared to sildenafil treated group (232.2), which is statistically significant difference ( $p < 0.05$ ). The pregnancy rates were 4 (16.7%) in group A and 11(73.3%) in group B which was statistically significant difference between the two groups ( $p < 0.05$ )

**Conclusions:** Both Sildenafil citrate and estradiol valerate may be used to improve endometrial receptivity, but this study showed that the pregnancy rate with Sildenafil was significantly higher than with estradiol.

**Keywords:** Infertility, Sildenafil, Estradiol valerate, Endometrial thickness endometrial receptivity

## Correspondence:

Sundus Ali Dawood

High Institute of Infertility Diagnosis and Assisted Reproductive Technologies, Al – Nahrain University, Baghdad, Iraq

E-mail: [raadreshan@yahoo.com](mailto:raadreshan@yahoo.com)

DOI: [10.31838/srp.2020.6.93](https://doi.org/10.31838/srp.2020.6.93)

©Advanced Scientific Research. All rights reserved

## INTRODUCTION

Infertility is a worldwide public health issue; infertility affects around 13 per cent of couples. Implantation failure occurs despite the efforts of fertility clinics in the field of assisted reproductive technology (ART); thus, the successful implantation requires high-quality embryos and adequate receptive endometrial development. [1](Chang et al., 2015)

Endometrial thickness is one of the best predictors of implantation. Numerous studies have shown that embryo implantation and clinical pregnancy rates (PRs) in patients with an endometrial thickness of  $> 9$  mm are significantly higher. [2,3] (Richter et al., 2007; Kovacs et al., 2003)

Thin endometrium, which is usually  $< 7$  mm, is supposed to be less able to sustain implantation and pregnancy.[4](Israel et al., 1996) so the minimum thickness needed for implantation is 7 mm [5]. (Zadehmodarres et al., 2017) also when ET is increased ( $> 14$ mm). there are significantly reduced in Implantation and pregnancy rates

The thickness of the endometrium depends on a variety of factors including reproductive age, menstrual cycle phase, concentration of ovarian hormones (estrogen and progesterone), and density of endometrial hormone receptors.[6] (Elnashar et al., 1995)

There are several causes of thin endometrium: permanent endometrial injury, decreased blood flow, endometrial resistance to estrogen, decreased blood flow, and testosterone overexposure.

There are few and mainly observational or experimental treatments for poor, non-responsive endometrium

including high doses of estrogens, piroxicam, HCG and granulocyte stimulating factor.[7] (Gleicher et al., 2011)

Treatments have also been proposed, such as acetylsalicylic acid. The result of the action is mixed. [8,9]( Hsieh et al., 2000; Hurst et al., 2005)

## Estrogen

The endometrium, being a hormone-dependent tissue, proliferates in response to estrogen that further induces the development of progesterone receptors. As a consequence, estradiol (E2) treatments were given to infertile patients who presented thin endometrium, in an effort to improve endometrial proliferation.

Most of the E2 treatment trials in patients with a thin endometrium dealt with frozen embryo transfer cycles. In addition, while several studies have been performed on estrogen therapy with various routes of administration, there are many routes and durations of administration of E2 including oral, transdermal, vaginal, and intramuscular. As regards pregnancy rates, no endometrial preparation procedure has a convincing advantage over another [10]. (Groenewoud et al., 2010)

Estrogen-induced endometrial proliferation primarily depends on the flow of blood to the basal endometrium.[11] (Sher & Fisch,2002)

The effect of estrogen on endometrium does not come from the neighboring ovaries through direct or regional transmission, rather than through the systemic circulation During controlled ovarian hyperstimulation cycles, Chen et al. analyzed the effect of extended estradiol valerate

administration and found that the average ET in the therapy group improved from 6,7 to 8,6 mm after estradiol valerate treatment from 14 to 82 days. In the research group the pregnancy rate was increased more than in the control group [12]. (Chen et al., 2006)

#### Sildenafil

In the treatment of recurrent implantation failure patient, vaginal sildenafil citrate has been attempted. Sildenafil citrate is a powerful and selective phosphodiesterase-5 (PDE-5) inhibitor responsible for cGMP degradation, thus increasing the vasodilatory impact of nitric oxide by preventing cGMP degradation, contributing to vascular relaxation and increased uterine blood flow and enhancing endometrial efficacy. This results in vascular relaxation and increased blood flow and enhanced endometrial thickness [13] (Fahmy et al., 2015)

Zinger et al. identified two Asherman's syndrome infertility patients who became pregnant after using 25 mg vaginal sildenafil citrate, four times a day for 6–14 days in the first half of their cycles [14]. (Zinger et al., 2006)

The implantation achieved depends on the ability of the blastocyst to invade the endometrium and establish blood supply, which requires the genes to generate the proteins required to digest the endometrial cell matrix, regulate cell development, and induce angiogenesis such as plasminogen activator inhibitor 1 (PAI-1), tumor suppressor factor (p53) and vascular endothelial growth factor (VEGF).

[15,16] (D Ly et al., 2010; Goodman et al., 2009)

Sildenafil citrate was substantially augmented in p53, and induced angiogenic responses with improved VEGF.

[17,18] (Di et al., 2010; Pyriochou et al., 2007)

Potential functional markers of endometrial receptivity, even promising, nevertheless are invasive and expensive. Therefore Transvaginal ultrasonography was suggested as an alternative method in endometrial receptivity assessment. It has been documented that endometrial thickness and pattern on the day before oocyte retrieval can be a predictor of pregnancy achievement.[19] (Naha & Khastgir, 2018).

This study to estimate the effect of estradiol (E2) vs sildenafil citrate on ultrasonographic endometrial thickness, pattern, and vascularity

## SUBJECTS AND METHODS

Ethical approval and consent to participate

The study was approved by the ethics committee of High Institute of Infertility Diagnosis and Assisted Reproductive Technologies/AI-Nahrain University, and informed consent was obtained from the patients for this study after explaining details and benefits as well as risks to them.

A comparative prospective study was done in IVF center of High Institute of Infertility Diagnosis and Assisted Reproductive Technologies/AI-Nahrain University/ Baghdad-Iraq over a period from July 2019 until March 2020 including 60 women with unexplained, primary or secondary infertility. The age ranges from 18-39 years old. The exclusion criteria were as follows: Patients aged more than 40 years, female who have any congenital uterine anomaly or acquired deformities of the uterine cavity that

interfere with embryo implantation (as Asherman Syndrome), Large intramural sub mucosal fibroid, severe endometriosis, female who have tubal factors that may interfere with conception (uni- or bilateral hydrosalpinx), thyroid disease, a female who has contraindications to estrogen treatment (e.g., history of stroke, DVT and Benign liver disease like Hepatocellular Adenomas), male factor infertility according to parameters of seminal fluid analysis.

Ultrasound examination and hysterosalpingography (HSG) was done for all patients to evaluate endometrium uterus, ovaries and patency of fallopian tubes.

All patients had undergone ovulation induction by one of these drugs (Letrozole, Clomid, Letrozole +Gonadotropins) depending on the age, hormonal status and previous cycle response. Patients with thin endometrium were randomly distributed in two groups. In group A, 30 patients were given estradiol valerate tablet (estrofem, Novo Nordisk) 2 mg every 12 hours, while in group B, 30 patients were included and were given sildenafil citrate 25 mg (Viagra, Pfizer Inc., New York, USA) vaginally every 6 hours from day 6 of the cycle. Follow up by serial trans-vaginal sonography was done, when at least one follicle reaching the size equal to or more than 17mm on cycle day 11-13, then the trigger was given urinary HCG (5,000IU, Pregnyl, Organon, Netherland), ultrasound study had been done for assessing both endometrial morphology (thickness and pattern), The maximum distance between the two endometrium – myometrium junction interfaces in the longitudinal plane of the uterus was described as the thickness of the endometrium. [20] (Fang et al., 2016). The number and size of Graafian follicles and also blood samples were taken from all patients for measuring the level of vascular endothelial growth factor in the serum. Then timed spontaneous coitus was recommended. 3 days later luteal phase was supported by vaginal progesterone (Cyclogest, Actavis, UK, 400mg). After 14 days  $\beta$  hCG test was done in blood to check biochemical pregnancy.

#### Statistical analysis

The collected data were coded, tabulated and statistically analyzed by Statistical Package for Social Sciences (SPSS) version 22.0. By applying appropriate statistical method data were presented as mean  $\pm$  SD. The groups were compared by the Student's paired t- test, chi square and ANOVA tests. The degree of association between continuous variables was calculated by Pearson's correlation coefficient (r). The level of significance was taken at P value < 0.05.

## RESULTS

Sixty patients had primary or secondary infertility, all patients had normal HSG, and all received ovulation induction treatment, and they all have no gynecological problem.

Demographic parameters and the basal hormonal profile Regarding the demographic parameters (age, BMI of patients, and duration of infertility) and the basal hormonal profile level (FSH, LH, PRL, E2, and TSH), the statistical analysis showed no significant differences (P>0.05) between studied groups. (Table 1).

Table 1: Demographic parameters and hormonal profile among the studied groups

parameters	Estradiol group N = (30) (Mean ± SD)	Sildenafil group N = (30) (Mean ± SD)	P value
Age (years)	28.49 ± 6.2	31.6 ± 5.6	0.084 <sup>NS</sup>
BMI (Kg/m <sup>2</sup> )	27.43 ± 1.98	28.49 ± 2.51	0.076 <sup>NS</sup>
Duration of infertility (years)	2.23 ± 1.56	3.07 ± 1.83	0.064 <sup>NS</sup>
FSH	6.29 ± 2.67	6.88 ± 2.82	0.41 <sup>NS</sup>
E2	44.31 ± 29.95	43.35 ± 12.02	0.87 <sup>NS</sup>
LH	6.63 ± 2.26	7.02 ± 2.81	0.56 <sup>NS</sup>
TSH	1.97 ± 1.07	2.29 ± 0.873	0.21 <sup>NS</sup>
PROLACTIN	7.49 ± 2.97	7.73 ± 2.90	0.75 <sup>NS</sup>

NS= insignificant ( $p > 0.05$ ), n=number, SD=standard deviation, BMI=body mass index, FSH=follicle stimulating hormone, LH=luteinized hormone, E2=estradiol hormone, PRL=prolactin hormone, TSH=thyroid stimulating hormone.

#### Ovulation induction protocols

Seventeen out of 60 patients in group A (28%) were stimulated using clomiphene citrate and 21 out of 60 (35%) were stimulated using Letrozole whereas rest 22 out of 60

(37% ) were given Letrozole and gonadotrophins (HMG/FSH). (Figure 1). None of these cycle characteristics had significant impact on the results obtained.

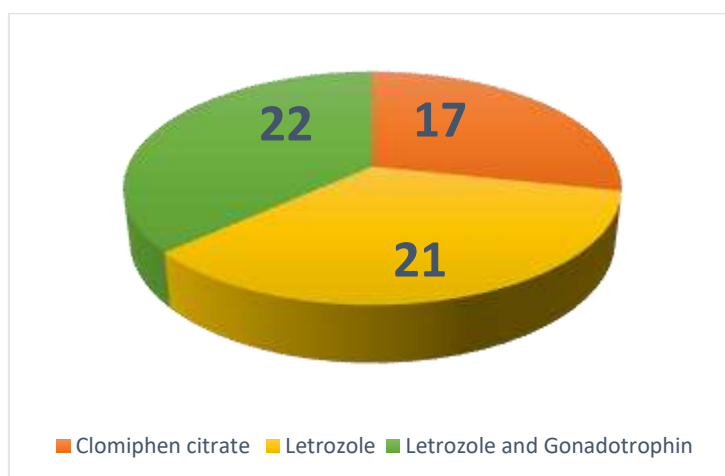


Figure 1: Distribution of patients according to OI treatment

Comparison between endometrial thickness at day 7 of the cycle and at day of HCG trigger

Figure (2) comparison between the ET at CD7 and ET at day of HCG trigger and the results demonstrated no

significant differences ( $P > 0.05$ ) between the two groups (A&B). On the other hand, there was statistically high significant difference between endometrial thickness at cycle day 7 and HCG trigger day within the groups ( $p < 0.001$ ).

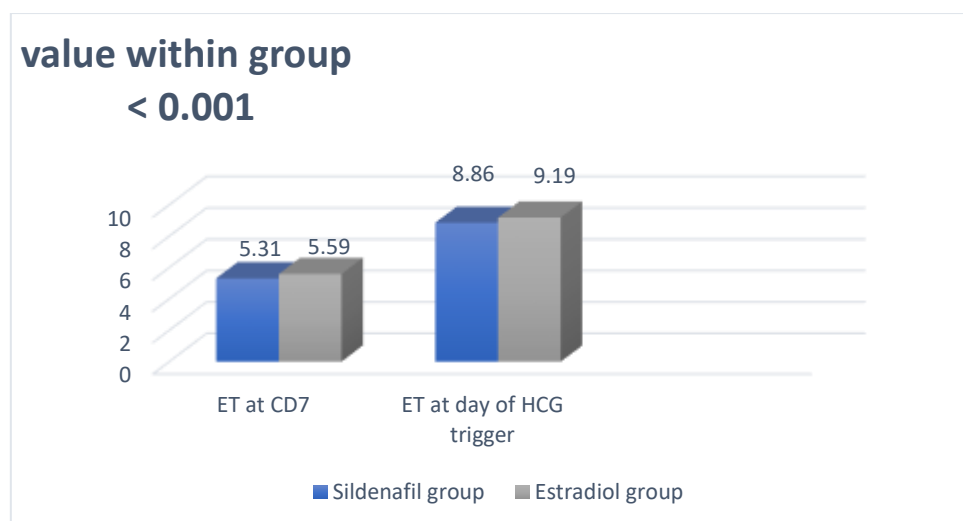


Figure 2: Comparison between endometrial thickness at day 7 of the cycle and at day of HCG trigger between and within groups

Ultrasound parameter and VEGF level at day of HCG administration

The difference in size and number of dominant follicles at HCG day between studied groups (A&B) was statistically insignificant ( $P>0.05$ ).

Mean endometrial thickness at day of HCG administration was 9.19 mm in estradiol treated group whereas 8.86 mm in sildenafil treated group. Although the endometrium was found to be thicker in estradiol treated group, the results were statistically insignificant ( $p > 0.05$ ).

The trilaminar pattern of endometrium was documented in (96.6%) of patients in group A and 90% in group B. There

were no significant differences between sildenafil and estradiol groups regarding the mean VEGF level which was 220.5, 232.2 respectively ( $p>0.05$ ).as showed in table 2

Pregnancy rates

The biochemical pregnancy test was positive in 4 females within group A (16.7%) and 11 in group B (73.3%). So a significant increase in biochemical pregnancy rates was also observed when sildenafil was used in patients with poor endometrium as compared to estradiol valerate group (Table 2).

Table 2: Comparison of outcomes between the studied groups

	Estradiol group N = (30) Mean $\pm$ SD	Sildenafil group N = (30) Mean $\pm$ SD	P value
ET at CD7 (mm)	5.59 $\pm$ 0.64	5.31 $\pm$ 0.69	0.115
ET at day of HCG (mm)	9.19 $\pm$ 0.657	8.86 $\pm$ 0.841	0.099
E. Pattern Triple layer	29 (96.6%)	27 (90%)	0.306
Mono layer	1 (3.3%)	3 (10%)	
Size of follicles at day of HCG (mm)	19.2 $\pm$ 1.57	19.41 $\pm$ 1.58	0.615
Number of follicles at day of HCG	1.53 $\pm$ 0.73	1.73 $\pm$ 0.64	0.264
VEGF level (pg./ml)	220.5 $\pm$ 19.05	232.2 $\pm$ 27.8	0.062
Pregnancy rate	4 (16.7%)	11 (73.3%)	0.037

\*Statistically significant ( $P<0.05$ ), n=number, SD=standard deviation, ET =endometrial thickness, HCG=human chorionic gonadotropin hormone, VEGF=vascular endothelial growth factor

## DISCUSSION

In the present study, the endometrial thickness and pattern was evaluated at the day of HCG trigger which was improved in both estradiol and sildenafil treatment. There was no significant different regard these parameters between studied groups however the pregnancy rate was significantly higher in sildenafil treated group

The results of the present study are in accordance with previous studies. In a comparative prospective study performed by Mangal that conclude: Mean endometrial

thickness at the day of HCG trigger are higher in estradiol group than in sildenafil group however Sildenafil when compared to estradiol valerate has better consequences to the grade that endometrial vascularity is concerned and improved pregnancy result.[21] Mangal and Mehiriishi,2016). While another study was done by Çetinkaya K who used estrogen vaginally in patients undergoing clomiphene citrate induced cycle. It showed substantial growth in endometrial thickness on ovulation day compared to the group where only clomiphene citrate was used but no improvement in pregnancy rate was

observed. [22] (Çetinkaya and Kadanalı, 2012.). Jimenez PT et al they reported appropriate development of endometrium in 67% patients when used oral estradiol 2 mg thrice daily from day one for 12 days. [23]. (Jimenez et al.,2013) In a meta-analysis performed by Torres RF et al, the use of pure ethinyl estradiol for thin endometrium treatment was investigated, the findings were: using ethinyl estradiol (EE) increases endometrial thickness compared to patients using only placebo [24] (Torres et al.,2005).

The findings of this research have been compared with those of previous research. In a retrospective study by Takasaki et al, used sildenafil intravaginal starting first day of cycle till day of ovulation and 92% patients showed improvement in endometrial thickness. [25] (Takasaki et al., 2010). Firouzabadi et al. conclude that endometrial thickness and triple-line pattern were significantly higher with sildenafil and estradiol than with estradiol valerate alone. Also, the rates of chemical pregnancy were higher but not statistically significant. [26] (Firouzabadi et al., 2014) . For a prospective study on fifty patients with unexplained 1ry infertility, El-Shourbagy et al were conclude that the patients treated with sildenafil citrate saw a statistically significant increase in endometrial thickness and a higher pregnancy rate than control group. [27] (El-Shourbagy et al., 2017). Vardhan et al. who observed similarly that use of Sildenafil citrate with Estradiol valerate substantially improves endometrial thickness compared to Estradiol valerate alone or control group. [28] (Vardhan et al., 2019). In his randomized placebo-controlled trial study, Moini et al. institutes that vaginal sildenafil is believably that the outcome of chemical pregnancy. two-fold increase compared to placebo in woman with recurrent IVF failure, but still no substantial difference in endometrial thickness between groups on the day of HCG injection [29] (Moini et al., 2020)

All of these studies further reinforce this research. Though most research compared estradiol or sildenafil alone with the patient's previous cycle taken as control. Several studies comparing to a combination of estradiol plus Sildenafil to estradiol alone have also been released.

## CONCLUSION

To sum up, an adequately thick, trilaminar endometrial pattern environment is essential for embryo implantation and continuation of pregnancy.

Both estradiol and sildenafil can be used to improve the endometrium (thickness and vascularity) and to support the development of embryos.

Sildenafil has substantially increased the outcome of pregnancy in patients undergoing a stimulated cycle relative to estradiol valerate, this can be explained by the increased vascular flow caused by enhanced VEGF production leading to increased endometrial growth and pattern leading to increased endometrial receptivity.

## FUNDING

No funding sources

## CONFLICT OF INTEREST

None declared

## REFERENCES

1. Chang, Y., Li, J., Chen, Y., Wei, L., Yang, X., Shi, Y., & Liang, X. (2015). Autologous platelet-rich plasma promotes endometrial growth and improves pregnancy outcome during in vitro fertilization. *International journal of clinical and experimental medicine*, 8(1), 1286.
2. Richter, K. S., Bugge, K. R., Bromer, J. G., & Levy, M. J. (2007). Relationship between endometrial thickness and embryo implantation, based on 1,294 cycles of in vitro fertilization with transfer of two blastocyst-stage embryos. *Fertility and sterility*, 87(1), 53-59.
3. Kovacs, P., Matyas, S. Z., Boda, K., & Kaali, S. G. (2003). The effect of endometrial thickness on IVF/ICSI outcome. *Human Reproduction*, 18(11), 2337-2341.
4. Israel, R., Isaacs, J. D., Wells, C. S., Williams, D. B., Odem, R. R., Gast, M. J., & Strickler, R. C. (1996). Endometrial thickness is a valid monitoring parameter in cycles of ovulation induction with menotropins alone. *Fertility and sterility*, 65(2), 262-266.
5. Zadehmodarres, S., Salehpour, S., Saharkhiz, N., & Nazari, L. (2017). Treatment of thin endometrium with autologous platelet-rich plasma: a pilot study. *JBRA assisted reproduction*, 21(1), 54.
6. Elnashar, A., Afifi, A., & Donia, O. (1995). Endometrial thickness and pregnancy rates in infertile couples undergoing AIH. *Benha MJ*, 12, 1-9.
7. Gleicher, N., Vidali, A., & Barad, D. H. (2011). Successful treatment of unresponsive thin endometrium. *Fertility and sterility*, 95(6), 2123-e13.
8. Hsieh, Y. Y., Tsai, H. D., Chang, C. C., Lo, H. Y., & Chen, C. L. (2000). Gynecology: low-dose aspirin for infertile women with thin endometrium receiving intrauterine insemination: a prospective, randomized study. *Journal of assisted reproduction and genetics*, 17(3), 174-177.
9. Hurst, B. S., Bhojwani, J. T., Marshburn, P. B., Papadakis, M. A., Loeb, T. A., & Matthews, M. L. (2005). Low-dose aspirin does not improve ovarian stimulation, endometrial response, or pregnancy rates for in vitro fertilization. *Journal of experimental & clinical assisted reproduction*, 2(1), 8.
10. Groenewoud, E. R., Cantineau, A. E., Kollen, B. J., Macklon, N. S., & Cohlen, B. J. (2017). What is the optimal means of preparing the endometrium in frozen-thawed embryo transfer cycles? A systematic review and meta-analysis. *Human reproduction update*, 23(2), 255-261.
11. Sher, G., & Fisch, J. D. (2002). Effect of vaginal sildenafil on the outcome of in vitro fertilization (IVF) after multiple IVF failures attributed to poor endometrial development. *Fertility and Sterility*, 78(5), 1073-1076.
12. Chen MJ, Yang JH, Peng FH et al (2006) Extended estrogen administration for women with thin endometrium in frozen-thawed in-vitro fertilization programs. *J Assist Reprod Genet*. 23:337-342



13. Fahmy, A. A., El Sakkary, M., & Sayed, S. (2015). The value of oral sildenafil in the treatment of female infertility: a randomized clinical trial. *Life Sci. J*, 12(4), 78-82.
14. Zinger, M., Liu, J. H., & Thomas, M. A. (2006). Successful use of vaginal sildenafil citrate in two infertility patients with Asherman's syndrome. *Journal of women's health*, 15(4), 442-444.
15. D Ly, K., Aziz, N., Safi, J., & Agarwal, A. (2010). Evidence-based management of infertile couples with repeated implantation failure following IVF. *Current Women's Health Reviews*, 6(3), 200-218.
16. Goodman, C., Jeyendran, R. S., & Coulam, C. B. (2009). P53 tumor suppressor factor, plasminogen activator inhibitor, and vascular endothelial growth factor gene polymorphisms and recurrent implantation failure. *Fertility and sterility*, 92(2), 494-498.
17. Di, X., Gennings, C., Bear, H. D., Graham, L. J., Sheth, C. M., White, K. L., & Gewirtz, D. A. (2010). Influence of the phosphodiesterase-5 inhibitor, sildenafil, on sensitivity to chemotherapy in breast tumor cells. *Breast cancer research and treatment*, 124(2), 349-360.
18. Pyriochou, A., Zhou, Z., Koika, V., Petrou, C., Cordopatis, P., Sessa, W. C., & Papapetropoulos, A. (2007). The phosphodiesterase 5 inhibitor sildenafil stimulates angiogenesis through a protein kinase G/MAPK pathway. *Journal of cellular physiology*, 211(1), 197-204.
19. Naha, M., & Khastgir, G. (2018). Tests for Endometrial Receptivity. *Practical Guide in Assisted Reproductive Technology*, 121.
20. Fang R, Cai L, Xiong F, Chen J, Yang W, Zhao X (2016) The effect of endometrial thickness on the day of hCG administration on pregnancy outcome in the first fresh IVF/ICSI cycle. *Gynecol Endocrinol* 32(6):473-476
21. Mangal, S., & Mehirishi, S. (2016). To study and compare the effect of vaginal sildenafil and estradiol valerate on endometrial thickness, blood flow and pregnancy rates in infertile women undergoing intrauterine insemination. *Int J Reprod Contracept Obstet Gynecol*, 5(7), 2274-2277.
22. Çetinkaya, K., & Kadanalı, S. (2012). The effect of administering vaginal estrogen to clomiphene citrate stimulated cycles on endometrial thickness and pregnancy rates in unexplained infertility. *Journal of the Turkish German Gynecological Association*, 13(3), 157.
23. Jimenez PT, Schon SB, Odem RR, Ratts VS, Jungheim ES. A retrospective cross-sectional study: fresh cycle endometrial thickness is a sensitive predictor of inadequate endometrial thickness in frozen embryo transfer cycles. *Reprod Biol Endocrinol*. 2013;11:35.
24. Torres, R. F., Habana, A. E., & Tansengco, L. G. (2005). The effect of estrogen supplementation on the endometrium and pregnancy rate among infertile women treated with clomifene citrate: a meta-analysis. *Fertility and Sterility*, 84, S162-S163.
25. Takasaki, A., Tamura, H., Miwa, I., Taketani, T., Shimamura, K., & Sugino, N. (2010). Endometrial growth and uterine blood flow: a pilot study for improving endometrial thickness in the patients with a thin endometrium. *Fertility and sterility*, 93(6), 1851-1858.
26. Firouzabadi, R. D., Davar, R., Hojjat, F., & Mahdavi, M. (2013). Effect of sildenafil citrate on endometrial preparation and outcome of frozen-thawed embryo transfer cycles: a randomized clinical trial. *Iranian journal of reproductive medicine*, 11(2), 151.
27. El-Shourbagy, S., Ossman, A. M., & El-Mohamady, A. (2017). The role of sildenafil citrate (viagra) suppositories on endometrial response (thickness and mean resistance index of endometrial spiral artery) in cases of unexplained infertility. *Menoufia Medical Journal*, 30(2), 343.
28. Vardhan, S., Yadav, P., Agarwal, R., Garg, R., Verma, U., & Pengoria, M. (2019). Effect of Sildenafil Citrate and Estradiol Valerate on Endometrial Characteristics in Ovulation-induced Cycle in Women with Dysovulatory Infertility. *Journal of South Asian Federation of Obstetrics and Gynaecology*, 11(3), 165.
29. Moini, A., Zafarani, F., Jahangiri, N., Sadatmahalleh, S. J., Sadeghi, M., Chehrizi, M., & Ahmadi, F. (2020). The Effect of Vaginal Sildenafil on The Outcome of Assisted Reproductive Technology Cycles in Patients with Repeated Implantation Failures: A Randomized Placebo-Controlled Trial. *International Journal of Fertility & Sterility*, 13(4), 289