Original Article

Comparison of Unstimulated In Vitro Maturation and Stimulated In Vitro Fertilization in Women with Poly Cystic Ovarian Syndrome

Z. Khashavi MD *, M.A. Karimzadeh MD *

ABSTRACT

Background: In regard to complications of controlled ovarian hyperstimulation (COH) such as multiple pregnancy and ovarian hyper stimulation syndrome (OHSS) and other short and long term complications, the interest to In Vitro Maturation (IVM) of oocytes from unstimulated ovaries is increasing. This clinical trial study was designed to establish the relative success of treatment with IVM in comparison with In Vitro Fertilization (IVF) in Poly Cystic Ovarian Syndrome (PCOS) women.

Methods: This prospective clinical trial included 35 IVM and 35 IVF cycles matched for age, and duration and cause of infertility. IVM patients underwent transvaginal retrieve of immature oocytes during unstimulated cycles, IVM and ICSI. Those in IVF group underwent ovarian stimulation after pituitary supression. Main outcome measures included number of retrieved oocytes, number of produced embryos, and pregnancy rate and complications.

Results: In the IVM group, 1.39 mature oocytes, 0.27 fertilized oocyte, and 0.24 embryos were obtained per retrieval, and in IVF group, 8.45 mature oocytes, 3.8 fertilized oocytes, and 2.7 embryos were obtained.

Conclusion: This significant differences between two groups (P~ 0) indicate that this method maybe used as a research setting until further study.

Key words: In Vitro Maturation, ART, unstimulated IVM, PCOS.

In vitro fertilization (IVF) is associated with two major complications: multiple pregnancy and OHSS due to controlled ovarian hyperstimulation (COH) especially in PCOS women. COH isn't required for In vitro maturation (IVM), Therefore, this method is cheaper and safer than IVF. In addition, regard to short and long term complications and side effects of COH, such as abdominal pain, nausea, and vomiting, unknown effects on dysplastic changes of ovaries [37], IVM of oocytes from unstimulated ovaries is a reproductive technology of increasing interest [35]. Indications for IVM include: PCOS women requiring IVF, primarily poor quality embryos in repeated previous IVF without apparent reason, poor responders to high dose gonadotrophin stimulation for IVF, women with PCOS who are considering egg donation, immature oocytes collected for ART, use of fresh or frozen ovarian tissues as sources of oocyte, IVM of germinal vesicle which is reconstructed [1].

In regards to different results of IVM in rates of oocyte maturation, fertilization, cleavage, and pregnancy [38,39], we designed this study to establish the relative success of unstimulated IVM in comparison with stimulated IVF in PCOS women.

Materials and Methods

During 2003, 70 PCOS women who were candidate for assisted conception were placed in two groups randomly: IVM and IVF. Finally, the prospective comparative study was done on 33 cycles of IVF.The patients commenced the treatment after full explanation and giving informed consent. An ovary was detected as PCO when showed ≥ 10 small (2-8 mm) cystic follicles around the periphery

* Reaserch and Cilinical Center for Infertility, Shahid Sadoughi University of Medical Sciences, Yazd, Iran.

of ovary with enlarge echogenic stroma, during early follicular phase, using transvaginal ulterasound [40]. Women with oligomenorrhea received IM injection of 100 mg of progestron to induce withdrawal bleeding. In IVM group, a baseline ultrasound was done for all women on cycle days 3 and 8 to exclude dominant follicle and ovarian cysts, because data suggest that the presence of dominant follicle at the time of oocyte retrieval is deleterious to outcome of IVM [12]. HCG priming, 10000 units, was done on days 9 to 12 [26] and oocyte retrieval was done 36h after hCG injection with 18 g single-lumen aspiration needle with aspiration pressure of 7.5 k_{Pa} under general anesthesia. Oocytes were collected in culture tubes containing Hams F10 with 2 Iu/ml of heparin. After washing of blood, immature oocytes were incubated in a culture dish containing TC-199 medium supplemented with 25 mol/lit of pyruvic acid, 75 mIu/ml of FSH+LH, and 10% FBS at 37°c, 5% co_2, and 95% air with high humidity. Maturity of oocytes was determined at 24 hours post collection denuded of cumulos. Mature oocytes were determined by presence of first polar body and ICSI was done and transferred into medium culture. Fertilization was assessed 18 hours after ICSI, with appearance of 2 pronuclei and 2 polarbodies. Embryo transfer was done on day 2 or 3 after ICSI. For endometrial preparation, Ethinyl estradiol was used depending on endometrial thickness on retrieval day and luteal support was provided by IM injection of 100 mg of progestron/day.

In IVF group, pituitary suppression was done by long protocol with superfact (buserline) and stimulation by hMG (225 unit/day), and serial control sonography was done to reveal mature follicles (at least 2 follicles with 18 mm in diameter). HCG priming with 10000 units of HCG was done and oocyte collection was done routinely and IVF or ICSI procedures were done traditionally. Outcome measure included the number of oocyte collected, metaphase II produced oocytes, embryos (2PN), pregnancy rate, and OHSS. In two groups statistical analysis were performed, using the SPSS statistical package. Chi square and fisher exact test were performed for statistical comparison between two groups.

Results

There were 35 women in IVM group and 35 in IVF. Two cycles in IVM group failed to reach egg retrieval. There were no differences in mean age, infertility cause, and previous IVF cycles. Oocytes retrieval in two groups were: 100% in IVF groups and 51.5% in IVM group (P~ 0); fertilization rate of mature oocytes in IVM group was 30% per cycle and 100% per cycle in IVF group (P~ 0); embryo transfer was 18.1% in IVM and 88.8% in IVF group (P~ 0); pregnancy rate was 3.3% in IVM and 18.8% in IVF groups (P~ 0); and OHSS was 11.2% in IVF group and 0% in IVM group (P<0.05) (Table 1).

Mean number of retrieved oocytes (mature or immature), fertilized oocytes, and transferred embryos in two groups showed significant differences (Table 2).

Table 1. Results of 33 unstimulated IVM and 35 stimulated IVF cycles in PCOS women.

	IVM	IVF	OR
Number of cycle	33	35	
Age	28.4	29.3	P= 0.05
Duration of infertility	8.2	7.5	P= 0.51
Oocyte collected (%)	17(51.5%)	35 (100%)	P= 0.000
Fertilized 2PN embryos (%)	10(30.3%)	35 (100%)	P= 0.000
Embryo transferred (%)	6(18.1%)	31(88.8%)	P= 0.000
Pregnancy rate (%)	1(3.03%)	4(11.4%)	P= 0.357
Moderation or sever OHSS (%)	0(0%)	4(11.4%)	

Table 2. Comparison of different outcome measures in two groups.

	IVM	IVF	P Value
Number of cycles	33	35	
Mean of retrieved oocytes per cycle	1.39	8.45	0.000
Mean of fertilized oocytes per cycle	0.27	3.85	0.000
Mean of transferred embryos per cycle	0.24	2.71	0.000

Table 3. Comparison of success rate in different stages cases with positive puncture in two groups.

Number	Positive puncture (percent)	Fertilized oocytes (percent)	Transfered embryos (percent)	Pregnancy rate (percent)
35	35 100%	35 100%	31 81%	4 (13%)
33	17 51%	10 60%	6 60%	1 16.6%
P value	0.000	0.000	0.03	61%

In table 3 we have compared the success rate in fertilization, and embryo transfering stages, and pregnancy rate in patients whom we succeed to retrieve oocytes.

Discussion

Laparoscopic electrocuagulation of the ovaries with PCOS can be an effective method for good response to stimulation, but adhesion formation results in 17% to 27% of patients. A similar outcome may be achieved by ovarian drilling by immature oocyte collection without adhesion. In addition, immature oocytes which retrieved by this method and used for IVM, can result in a good embryos.

Recent papers showed a high incidence (21%) of non cleavage embryos produced by IVM, and further (27%) failed to develop beyound the first division. In addition, the remaining embryos showed high rates of aneuploidy. Maybe, specific time and mediators affect from circulation and other follicles or cells around the immature oocyte came to expression or imprinting specific gene in vivo [31, 34]. Therefore, it would be unwise to use these embryos in clinical setting until further research. Early embryo development was controlled by maternal genome which inheritated by oocyte genome. Therefore, gene expression and imprinting during oocyte maturation stages affect on embryo development [40]. Both nuclear and cytoplasmic maturation need to ensure developmental competence of embryos. Synchrony between endometrial and embryonic development in fresh IVM ET isn't ideal [41]. The results of this study demonstrate that immature oocytes from PCOS patients could mature and fertilize invitro. Although number of oocytes which have been retrieved and number of embryos produced in this method were lower than other studies reported by Trounson et al [9], Barnes et al [20], TJ Child [2], and Tan SL [1], but development in culture system and experience in oocyte retrieval from small follicles may provide an advantage for assisted reproductive in PCOS women in our center.

In regards to other studies [19,1,2] in spite of our study, this protocol is a useful alternative treatment for PCOS women who are the candidates for ART.

References

1. *Wittemore AS. Fertility drugs and the risk of ovarian cancer. Hum Reprod 2000; 15: 1685-90.*
2. *Cha KY, Han SY, Chung HM, et al. Pregnancies and deliveries after IVM culture followed by IVF and Et without stimulation in PCO. Fertil Steril 2000; 73: 978-83.*
3. *Trounson A, Wood C, Kausche A. IVM and fertilization and developmental competence of oocytes recovered from unstimulated PCO patients. Fertil Steril 1994; 62: 353-62.*
4. *Cha KY, Chain RC. Maturation in Vitro of immature human oocytes for clinical use. Hum Reprod update 1998; 4: 103-20.*
5. *Moor RM, Dai Y, Lee C. Oocyte maturation and embryonic failure. Hum Reprod update 1998; 4: 223-36.*
6. *Russell JB. Immature oocyte retrieval combined with IVM. Hum. Reprod 1998; 13 suppl 3: 63-70.*
7. *Mikkelsen AL, Smith SD, Lindenberg S. Human oocytes from regularly menstruating women. Hum Reprod 1999; 14: 1847-51.*
8. *Sutton ML, Gilchrist RB, Thompson JG. Effects of in-vivo and in-vitro environments on the metabolism of the cumulus-oocyte complex and its influence on oocyte developmental capacity. Hum Reprod Update 2003 Jan-Feb;9(1):35-48.*
9. *Suikkari AM, Tulppala M, Tuuri T, Hovatta O, Barnes F. Luteal phase start of low-dose FSH priming of follicles results in an efficient recovery, maturation and fertilization of immature human oocytes. Hum Reprod 2000 Apr;15(4):747-51.*
10. *Trouson A, Wood C, Kausche A. IVM and fertilization and developmental competence of oocytes of unstimulated PCOS patients. Fertil Steril 1994; 62: 353-62.*
11. *Barnes FL, Kausche A, Tiglias J, Wood C, Wilton L, Trounson A. Production of embryos from in vitro-matured primary human oocytes. Fertil Steril 1996 Jun;65(6):1151-6.*
12. *Child TJ, Phillips SJ, Abdul-Jalil AK, Gulekli B, Tan SL. A comparison of in vitro maturation and in vitro fertilization for women with polycystic ovaries. Obstet Gynecol 2002 Oct;100(4):665-70.*
13. *Tan SL, Child TJ. In-Vitro maturation of oocytes from unstimulated polycystic ovaries. Reprod Biomed online 2002; 4 suppl 1: 18-23.*
14. *Barnes FL, Crombie A, Gardner DK, Kausche A, Lacham-Kaplan O, Suikkari AM, et al. Blastocyst development and birth after in-vitro maturation of human primary oocytes, intracytoplasmic sperm injection and assisted hatching. Hum Reprod 1995 Dec;10(12):3243-7.*