Artificial endometrial preparation for oocyte donation using synthetic estrogen and progestogen

A. Abu-Musa, A. Hannoun, A. Khalil, Z. Masaad, K. Karam

Department of Obstetrics and Gynecology, American University of Beirut - Medical Center, Beirut (Lebanon)

Summary

Objective: To evaluate the outcome of an oocyte donation program using synthetic estrogen and progestational agents for uterine preparation.

Methods: Conjugated estrogen, 1.25 mg per day at increasing doses and dydrogesterone, a synthetic progestogen, 30 mg per day were used for uterine priming. All embryo transfers were done on day 2 of progestogen supplementation.

Results: The pregnancy rates were 38% per embryo transfer. The ongoing pregnancy rate was 31% with an abortion rate of 20%.

Conclusion: Endometrial preparation in an oocyte donation program using orally administered synthetic estrogen and progestogen gives pregnancy rates comparable to those reported with natural products.

Key words: Oocyte donation; Conjugated estrogen; Dydrogesterone.

Introduction

Implantation remains to be the major factor restricting the success of assisted conception. This crucial process is still incompletely understood. The implantation window, the period during which the endometrium is receptive for implantation, is affected by the sequential action of estrogen and progesterone. Oocyte donation is a good model for understanding oocyte and endometrial function in reproduction [1]. Several protocols for uterine preparation using estradiol and progesterone have been suggested. In almost all protocols the most commonly used estrogen is the micronized 17 ß-estradiol and estradiol valerate, and the most commonly used progesterone is intramuscular oil and micronized progesterone. In addition, the highest pregnancy rate is achieved in embryos transferred to recipients on days 3 to 5 of progesterone supplementation. In this study we report our preliminary experience in oocyte donation using conjugated estrogen and synthetic progestogen for uterine priming and performing early embryo transfer on day 2 of progestogen supplementation.

Material and Methods

Donors:

Oocyte donors were 27 patients undergoing induction for assisted reproduction who agreed to donate their excess oocytes anonymously. They were aged ≤ 32 years, with a mean age ± SD of 27.92 ± 2.65 (range 23 to 32 years). The stimulation protocol in all donors included administration of 900 µg/day buserelin acetate nasal spray (Suprefact; Hoechst AG, Frankfurt, Germany) from day 21 of the previous cycle to the day of human chorionic gonadotrophin (hCG) administration. Four ampoules of human menopausal gondotropins (hMG; Humegon, Organon Ltd, Oss, The Netherlands) were administered daily as of the third day of the menstrual cycle. Patients were monitored by serum estradiol levels and transvaginal ultrasound scans. When two follicles reached a mean diameter of 18 mm, 10,000 IU hCG (Pregnyl; Organon Ltd) were given. Oocyte retrieval was scheduled 34-36 hours after hCG administration using an ultrasound-guided transvaginal approach.

Recipients:

A total of 35 patients underwent 52 cycles of oocyte donation. All patients had early menopause or premature ovarian failure. Patients on stand-by to receive donated ova were treated with the same protocol for endometrial preparation. Conjugated estrogen (Premarin; Wyeth-Ayrest Laboratories, Montreal, Canada) was started immediately upon admission to the treatment cycle. Premarin 1.25 mg/day was given for four days, 2.5 mg/day for another four days, and 3.75 mg/day thereafter. The duration of treatment with 3.75 mg/day of conjugated estrogen varied in accordance with availability of the oocytes but not exceeding 24 days.

On the day of recovery of the donated oocytes, a progestogen supplement as 30 mg/day of dydrogesterone (Duphaston, Solvay Duphar, Weesp, The Netherlands) was started. Donated oocytes were distributed to one or more recipient, each receiving 4 to 6 oocytes. Oocytes were inseminated and cultured in human tubal fluid medium (HTF; Irvine Scientific, Santa Ana, California, U.S.A.) supplemented with 0.5% bovine serum albumin (Irvine Scientific). Embryo transfer up to 5 embryos was performed 48 hours following oocyte recovery, i.e., on day 2 of progestogen supplementation. Fourteen days later, a pregnancy test was performed and if positive, the daily doses of conjugated estrogen and dydrogesterone were continued until 12 weeks of gestation.

Results

Thirty-five patients underwent a total of 52 embryo transfers. Eighteen patients got pregnant, one with a twin

Received May 5, 1998
revised manuscript accepted for publication June 9, 1998

gestation and 2 patients got pregnant twice. Our pregnancy rate per embryo transfer was 38% (20 pregnancies out of 52 embryo transfers). Four patients ended in spontaneous abortion at 7-10 weeks of gestation (abortion rate 20%), with 16 remaining on-going pregnancies (31% per embryo transfer).

Discussion

Since egg donation has become a well established technique, major questions arise concerning the optimal mode of hormonal replacement therapy and the duration of the receptive phase of the uterus, the so called "implantation window". Success of an in vitro fertilization cycle depends on oocyte quality, endometrial receptivity and their synchronization [2-4]. However, to-date, implantation phenomena have not been fully understood. The endometrium which is responsible for providing an environment receptive to the implantations is under steroid control. However, little is known about the hormonal requirements for implantation, although it is thought that receptivity is dependent on adequate secretory change induced by progesterone on a favorable proliferative endometrium primed by estrogen [5].

Many hormone replacement protocols have been developed [6-9]. The most commonly used estrogen is orally administered estradiol valerate and the most commonly used progesterone is a natural preparation in the form of oil injections or oral micronized forms. In this study, endometrial preparation was done using synthetic estrogen and progestogen. Premarin, a conjugated estrogen, is the most widely used preparation for oral estrogen replacement therapy. The reason we used premarin for endometrial preparation was simply the unavailability of natural estrogen. Although the safety of the use of non-human hormones in women attempting to conceive might be questionable, other studies that have used premarin in oocyte donation did not show any deleterious effects to the medication [6]. In addition, estradiol valerate, micronized estradiol or conjugated estrogens have been used without any obvious advantage of one drug over another [10]. Although premarin is a combination of up to 10 different estrogenic compounds, a significant proportion of its observed biologic response is thought to be mediated through estradiol regenerated in the liver from the reservoir of circulating sulfated estrogen [11]. Estradiol, a natural estrogen, is the most commonly used drug in oocyte donation. The relative potency of estradiol has been evaluated in several different studies. Gonadotropin suppression data suggest that estradiol is slightly less potent than conjugated estrogen [2]. The reason we chose the 1.25 mg of premarin at increasing doses is because the therapeutic equivalence of 1 mg of estradiol is 0.625 mg of conjugated estrogen [13]. Because it was our preliminary experience in oocyte donation, the step-up protocol was used. However, recently continuous single doses of estrogen are estrogen are being used. The duration of estrogen treatment varied between 11 to 24 days. The effect of the duration of premarin replacement was not evaluated in this study because several studies have shown that it has no effect on pregnancy rates [1, 14].

The progestogen used in our study was dydrogesterone. To our knowledge, this is the first report that uses dydrogesterone in a donor-recipient program. Dydrogesterone is an oral progesteron whose molecular structure closely resembles that of natural progesterone. We chose this compound because it an orally active progestogen which is indicated for use in pregnancy such as for the treatment of threatened abortion and recurrent early pregnancy loss with no specific contraindications. It is not a derivative of the 19-nortestosterone group which has been associated with androgenic effects on the developing female fetus. Yovich and Lower have used medroxyprogesterone as the support progesterone in patients with recurrent miscarriage and consider it to be safe [15]. In this study, no endometrial biopsies were done to evaluate the effect of dydrogesterone on the endometrium. However, previous studies have shown that Duphaston 10 mg/day for 10 days was a highly potent orally active progestational agent which induced complete secretory transformation of the endometrium and a histological appearance identical to that seen in natural cycles [16, 17]. In addition, there was no discrepancy between the extent of glandular and stromal reaction [17]. The pregnancy rate achieved in this study and which is comparable to other studies also suggests that the endometrial preparation and luteal support were adequate [1, 6, 18]. The most popular approaches today for endometrial preparation in oocyte donation cycles remain the vaginal or intramuscular routes. However, the local pain at the injection site is troublesome, especially in prolonged oocyte donation treatment. On the other hand, progesterone suppositories are associated with a discharge that is unacceptable to many patients. Therefore, one can see the possible advantage of the oral route especially with respect to patient compliance.

In this study all embryo transfers were done 48 hours after starting dydrogesterone supplementation. Previous studies suggest that in order to optimize success with IVF in oocyte recipients, embryo transfer should be performed on days 3-5 of progesterone supplementation [19-21]. Based on our results it seems that performing embryo transfers on day 2 of progestogen supplementation in donor-recipient cycles would give adequate pregnancy rates.

In conclusion, endometrial preparation in an oocyte donation program using orally administered synthetic estrogen and progestogen gives an adequate pregnancy rate comparable to that reported with natural products. The use of oral progestogen would make the treatment protocol simpler.

References

[1] Navot D., Bergh P. A., Williams M., Garrisi G. J., Guzman I., Sandler B. *et al.*: "An insight into early reproductive processes through the in vivo model of ovum donation". *J. Clin. Endocrinol. Metab.*, 1991, *72*, 408.

[2] Levran D., Dor J., Rudak E., Nebel L., Ben-Shlomo I., Ben Rafael Z. *et al.*: "Pregnancy potential of human oocytes - the effect of cryopreservation". *N. Engl. J. Med.*, 1990, *323*, 1153.
[3] Paulson R. J., Sauer M. V., Lobo R. A.: "Embryo implantation after human in vitro fertilization: importance of endometrial receptivity". *Fertil. Steril.*, 1990, *53*, 870.
[4] Chang M. C.: "Development and fate of transferred rabbit ova or blastocyst in relation to the ovulatory time of recipients". *J. Exp. Zool.*, 1950, *114*, 197.
[5] Serhal P. F., Craft I. L.: "Ovum donation - a simplified approach". *Fertil. Steril.*, 1987, *48*, 265.
[6] Ben-Nun I., Ghetler Y., Gruber A., Jaffe R., Fejgin M.: "Egg donation in an in vitro fertilization program: an alternative approach to cycle synchronization and timing of embryo transfer". *Fertil. Steril.*, 1989, *52*, 683.
[7] De Ziegler D., Frydman R.: "Different implantation rates after transfers of cryopreserved embryos originating from donated oocytes or regular in vitro fertilization". *Fertil. Steril.*, 1990, *54*, 682.
[8] Navot D., Bergh P. A., Williams M., Garrisi G. J., Guzman I., Sandler I. *et al.*: "Poor oocyte quality rather than implantation failure as a cause of age related decline in female fertility". *Lancet*, 1991, *337*, 1375.
[9] Sauer M. V., Paulson R. J., Macaso T. M., Francis M. M., Lobo R. A.: "Oocyte and pre-embryo donation to women with ovarian failure: an extended clinical trial". *Fertil. Steril.*, 1991, *55*, 39.
[10] Younis S. J., Simon A., Laufer N.: "Endometrial preparation: lesson from oocyte donation". *Fertil. Steril.*, 1996, *66*, 873.
[11] Barnes R. B., Lobo R. A.: "Pharmacology of estrogens". In: Mishell D. R. (ed.): "Menopause: physiology and pharmacology". Chicago, IL: Year Book, *Medical*, 1987, *301*, 315.
[12] Mashchak C. A., Lobo R. A., Dozono-Takano R., Eggena P., Nakamura R. N., Brenner P. F. *et al.*: "Comparison of pharmacodynamic properties of various estrogen formulations". *Am. J. Obstet. Gynecol.*, 1982, *144*, 511.
[13] Archer D. F., Fischer L. A., Rich D.: "Estrace vs premarin for treatment of menopausal symptoms: dosage comparison study". *Adv. Ther.*, 1992, *9*, 21.
[14] Remohi J., Gallardo E., Guanes P. P., Simon C., Pellicer A.: "Donor-recipient synchronization and the use of gonadotrophin releasing hormone agonist to avoid the premature luteinizing hormone surge in oocyte donation". In: Simon C. and Pellicer A. (eds.): "Regulators of Human Implantation". Oxford University Press, Oxford, 1995, 84.
[15] Yovich J., Lower A.: "Implantation failure: clinical aspects". *Baillieres Clin. Obstet. Gynaecol.*, 1991, *5*, 211.
[16] Tillinger K. G., Diczfalusy E.: "Progestational activity of stereoisomeric progesterone-analogues following oral administration in amenorrhea". *Acta Endocrinol.*, 1962, *35*, 197.
[17] Bishop P. M. F., Borell U., Diczfalsy E., Tillinger K. G.: "Effect of dydrogesterone on human endometrium and ovarian activity". *Acta Endocrinol.*, 1962, *40*, 203.
[18] Younis J. S., Mordel N., Lewin A., Simon A., Schenker J. G., Laufer N.: "Artificial endometrial preparation for oocyte donation: the effect of estrogen stimulation on clinical outcome". *J. Assist. Reprod. Genet.*, 1992, *6*, 222.
[19] Navot D., Laufer N., Kopolovic J., Rabinowitz R., Birkenfeld A., Lewin A. *et al.*: "Artificial induced endometrial cycles and establishment of pregnancies in the absence of ovaries". *N. Engl. J. Med.*, 1986, *314*, 806.
[20] Navot D., Scott R. T., Droesch K., Veeck L. L., Liu H-C., Rosenwaks Z.: "The window of embryo transfer and efficiency of human conception in vitro". *Fertil. Steril.*, 1991, *55*, 114.
[21] Rozenwaks Z.: "Donor eggs: their application in modern reproductive technologies". *Fertil. Steril.*, 1987, *47*, 895.

Address reprint requests to:
Dr. ALI KHALIL
Department of Obstetrics and Gynecology
American University of Beirut - Medical Center
P.O. Box: 113-6044/C8
Beirut (Lebanon)