Impact of Varying Stages of Endometriosis on the Outcome of In Vitro Fertilization–Embryo Transfer

LUBNA PAL,[1,3] JAN L. SHIFREN,[2] KEITH B. ISAACSON,[1] YuCHIAO CHANG,[2] LUCY LEYKIN,[1] and THOMAS L. TOTH[1]

Submitted: June 18, 1997
Accepted: August 7, 1997

Purpose: *The impact of severity of endometriosis on the outcome of in vitro fertilization (IVF) was analyzed in an uncontrolled, retrospective study in an academic IVF program.*
Methods: *Sixty-one patients with a primary diagnosis of endometriosis undergoing 85 cycles of IVF were included in the study. Patients were divided according to the severity of disease based on the revised American Fertility Society (AFS) classification into groups A (stages I/II, or minimal/mild) and B (stages III/IV, or moderate/severe). Group A included 32 patients undergoing 45 IVF–embryo transfer (ET) cycles; group B included 29 patients undergoing 40 IVF cycles. Exclusion criteria were age older than 40 years, basal day 3 follicle stimulating hormone (FSH) greater than 20 IU/L, male-factor infertility, assisted hatching, and gamete intrafallopian transfer cases. Stimulation for IVF cycles was standard using pituitary down-regulation with gonadotropin-releasing hormone agonist in a midluteal protocol. Controlled ovarian hyperstimulation (COH) was achieved using a combination of FSH and human menopausal gonadotropin. Outcomes assessed included response to COH and number, maturity, and quality of oocytes retrieved. Fertilization, implantation, and pregnancy rates after IVF-ET were also analyzed.*
Results: *The response to COH and the number, maturity, and quality of the oocytes was comparable between patients with varying severity of endometriosis. Fertilization rates for oocytes of patients in group B (stages III/IV) were significantly impaired compared to those in group A (stages I/II) ($P = 0.004$). The rates for implantation, clinical pregnancy, and miscarriage were comparable between the two groups.*
Conclusions: *The reduced fertilization potential of the oocytes obtained from patients with severe endometriosis in the absence of male-factor infertility suggests an adverse biological impact of the advanced disease on the oocytes. The outcome of IVF-ET, however, is unaffected by increasing severity of endometriosis. This suggests that IVF may compensate for or overcome this reduction in the biological potential of the oocytes associated with severe disease, thus accounting for a comparable outcome irrespective of the severity of endometriosis.*

KEY WORDS: endometriosis; staging; American Fertility Society classification; in vitro fertilization.

INTRODUCTION

Myriad hypotheses have been propounded in an attempt to explain the subfertility associated with endometriosis. Severe degrees of endometriosis (1) are known to induce anatomical distortions interfering with the timely union of the gametes. Pathophysiology of subfertility seen with stages I/II of the disease remains obscure. Over the past decade, infertility associated with endometriosis has been an expanding indication for pursuing in vitro fertilization (IVF). Of the available data regarding the impact of varying degrees of endometriosis on the outcome of IVF, conclusions have ranged from impaired outcome with increasing severity (2–5) to comparable results with different stages (6–8). One study has even shown an improved outcome of IVF with increasing severity of the disease (9). A number of these studies have not been well controlled for factors influencing the outcome of assisted reproductive techniques and the criteria for patient inclusion have been fairly broad, including male-factor infertility (2,3,10). Furthermore, most of the groups have failed to control for the day 3 follicle

[1] Departments of Reproductive Endocrinology and Infertility, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts 02114.
[2] Medical Practices Evaluation Center, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts 02114.
[3] To whom correspondence should be addressed at Massachusetts General Hospital, Vincent I Room 137 G, Boston, Massachusetts 02114.

1058-0468/98/0100-0027$15.00/0

stimulating hormone (FSH) and estradiol (E_2) levels, which are known independent predictors of outcome of IVF.

The aim of the present study was to analyze the outcome of IVF in patients with a primary diagnosis of endometriosis as a cause for their infertility, thereby enabling better counseling of patients with varying stages of the disease. We have tried to use stringent criteria for inclusion in an attempt to minimize an introduction of bias. Parameters assessed include the response to controlled ovarian hyperstimulation (COH), rates of fertilization, abnormal fertilization, cleavage and pregnancy rates, and rates of pregnancy loss including chemical pregnancies and clinical miscarriages for patients with varying stages of endometriosis.

MATERIALS AND METHODS

The medical records of all patients undergoing IVF in the program at Massachusetts General Hospital between January 1994 and April 1997 were reviewed. A total of 61 patients met the inclusion criteria and underwent 85 cycles of IVF. All the patients had a laparoscopically proven diagnosis of endometriosis as the primary cause of infertility. The severity of the disease was categorized according to the revised American Fertility Society (AFS) classification (1). The patients were divided into two groups based on the stage of endometriosis: patients with stages I and II (Group A) and those with stages III and IV (Group B) of the disease. Exclusion criteria were established, and the corresponding numbers of patients excluded from the study are as follows: age older than 40 years (6 patients); day 3 FSH of 20 mIU/ml or greater (17 patients; FSH less than 20 mIU/ml was considered normal for day 3 of the menstrual cycle in the assay used); IVF cycles using GnRH agonist in a nonluteal protocol (2 patients); male-factor infertility, defined as a concentration of motile sperm less than 10 million/ml and sperm with normal morphology less than 4% using strict criteria (27 patients); gamete intrafallopian transfer (GIFT) procedure (4 patients); and assisted hatching (6 patients).

The IVF protocol used pituitary down-regulation using the gonadotropin-releasing hormone agonist (GnRH-a) leuprolide acetate (LA; Lupron, TAP Pharmaceuticals, Deerfield, Illinois) commencing in the mid-luteal phase of the preceding cycle. COH with human menopausal gonadotropin (hMG); Humegon; Organon, West Orange, New Jersey) and/or FSH (Metrodin; Serono Laboratories, Randolph, Massachusetts) was initiated after confirmation of adequate pituitary suppression and the gonadotropins were continued in an individual step-down protocol. Ovulation was triggered with 10,000 IU human chorionic gonadotropin (hCG; Profasi; Serono Laboratories) when at least three follicles of 16 mm or greater were noted on serial ultrasound monitoring. Transvaginal ultrasound-guided oocyte retrieval followed approximately 35–36 hr after hCG administration. Oocytes retrieved were graded in terms of maturity based on the morphological appearance of the oocyte–cumulus complex. The embryos were graded according to the symmetry of the blastomeres and the presence or absence of fragmentation (grades 1–3), grade 1 being the best quality. Transcervical intrauterine embryo transfer (ET) was performed 48 or 72 hr following retrieval. Luteal-phase support was provided with intramuscular progesterone, 50 mg daily. Quantitative estimation of serum βhCG was performed 14 days after ET. A serum level of greater than 5 IU/L was considered as positive. A chemical pregnancy was said to occur if the initial serum βhCG level was greater than 5 IU/L but subsequently did not escalate and no intrauterine gestation sac was identified on transvaginal scan (TVS). A clinical pregnancy was defined as identification of an intrauterine sac(s) on TVS. Any pregnancy continuing beyond 20 weeks of gestation was considered as ongoing. Miscarriage was defined as a loss of a clinical pregnancy prior to 20 weeks.

Data were analyzed by performing a univariate analysis using the two-tailed Student's *t* test and Fisher's exact test, as appropriate. A multivariate regression analysis model was created to identify variables correlating with the outcome of IVF cycles. A logistic regression model controlled for covariates other than the stage of endometriosis. A stepwise procedure was used when the significance levels for entering and staying at the model were 0.3 and 0.05, respectively. The terms included in the model selection were the age; total ampoules; day of hCG; number of quality 1, 2, and 3 oocytes; fertilization rate; abnormal fertilization rate; and cleavage rate. The stage of endometriosis was never included in the model at any stage of the selection process. The SAS statistical package (SAS Institute, Cary, NC) was used for data analysis. Statistical significance was defined as P less than 0.05.

RESULTS

The data from 61 patients corresponding to 85 IVF cycles were analyzed and are shown in Tables I–IV.

Table I. Patient Characteristics in Groups A (Stages I/II) and B (Stages III/IV)[a]

	Group A	Group B	*P* value
Number of patients	32	29	NS
Age (years)	33.6 ± 3.0	34.4 ± 4.0	NS
Basal day 3 FSH (mIU/ml)	13.0 ± 4.0	13.4 ± 3.0	NS
Basal day 3 E_2 (pg/ml)	67.4 ± 36.3	64.9 ± 25.1	NS

[a] Values are means ± SD.

Table II. Response to COH in Groups A (Stages I/II) and B (Stages III/IV)[a]

	Group A	Group B	*P* value
Cycles of COH for IVF	45	40	NS
Total ampoules/cycle	38.1 ± 15.1	46.5 ± 19.3	0.029*
Day of hCG	11.6 ± 1.4	12.0 ± 1.8	NS
Max. E_2 on day of hCG (pg/ml)	2123 ± 1088	1838 ± 850	NS
Endometrial thickness on day of hCG (mm)	9.6 ± 1.7	10.1 ± 2.9	NS

[a] Values are means ± SD.
* Statistically significant.

The age and day 3 serum FSH and E_2 levels were comparable in the two groups (Table I). The outcome of COH was comparable in patients with varying severities of endometriosis (Table II). Comparable numbers of oocytes including preovulatory, immature, and postmature oocytes were recovered from patients in the two groups (Table III). The patients in group B required an excess of ampoules of gonadotropins to attain a serum E_2 comparable to that of group A (Table II), the difference between the groups being significant ($P < 0.05$). Analyzing the outcome, the only significant difference between the IVF cycles in the two groups was the impaired fertilization rate exhibited by oocytes retrieved from patients with moderate/severe disease (group B) (Table IV). All the cycles in group A ended in ET. One of the 40 IVF cycles in group B did not have an ET secondary to abnormal fertilization; this IVF cycle was included in the analysis of response to COH but excluded from assessment of the outcome of IVF in the severe group. The quality and number of embryos transferred were comparable between the two groups of patients. The total pregnancy rate and the chemical and clinical pregnancy rates were comparable between patients within the two groups, as was the rate of clinical miscarriage (Table IV).

Table III. Oocyte Characteristics for Groups A (Stages I/II) and B (Stages III/IV)[a]

	Group A	Group B	*P* value
Oocytes per cycle	11.2 ± 5.0	10.5 ± 5.0	NS
Mature oocytes per cycle	9.4 ± 4.2	9 ± 4.3	NS
Immature oocytes per cycle	1.4 ± 2.0	1.2 ± 2.0	NS
Postmature oocytes per cycle	0.4 ± 1.0	0.3 ± 1.0	NS
Quality 1 oocytes per cycle	1.0 ± 2.0	2.7 ± 4.0	NS
Quality 2 oocytes per cycle	6.0 ± 4.0	4.9 ± 3.0	NS
Quality 3 oocytes per cycle	3.0 ± 2.5	2.8 ± 2.0	NS

[a] Values are means ± SD.

Table IV. Outcome of IVF in Groups A (Stages I/II) and B (Stages III/IV)

	Group A	Group B	*P* value
Fertilization rate (%)[a]	81.3 ± 18	68 ± 24	0.004*
Abnormal fertilization rate (%)[a]	14.3 ± 16	19.5 ± 25	NS
Cleavage rate (%)[a]	93.3 ± 15	88 ± 20	NS
Embryos transferred per cycle[a]	4.0 ± 1.0	4.0 ± 1.0	NS
Implantation rate	19% (34/179)	15.4% (24/156)	NS
Total pregnancy rate per transfer	62.2% (28/45)	54% (21/39)	NS
Clinical pregnancy rate per transfer	51.1% (23/45)	38.5% (15/39)	NS
Chemical pregnancy rate per transfer	11.1% (5/45)	15% (6/39)	NS
Clinical miscarriage rate	13% (3/23)	13% (2/15)	NS
Ongoing pregnancy rate per transfer	44.0% (20/45)	33.0% (13/39)	NS

[a] Values are means ± SD.
* Statistically significant.

Univariate and multivariate analyses were undertaken to analyze the differences between successful and failed IVF cycles in the patients included in the study. The stage of endometriosis was not shown to be an independent predictor of the outcome of the IVF-ET cycle.

DISCUSSION

The data presented clearly demonstrate that the stage of endometriosis does not affect the outcome of IVF, a finding consistent with other studies published recently (6–8). We have shown a comparable response to COH, similar implantation rates, and similar rates of clinical and ongoing pregnancies. The rate of pregnancy losses, including chemical pregnancies as well as clinical miscarriages, was no higher in patients with severe endometriosis as suggested earlier by some groups. We have, furthermore, demonstrated that the quality of oocytes was not adversely affected by increasing sever-

ity of the disease. Earlier assisted reproduction technology programs used a laparoscopic approach for retrieval of oocytes (4,11,12) and their experiences suggested a poor outcome of IVF-ET with increasing severity of endometriosis. The technique of aspiration using the TVS-guided approach has yielded a comparable number of oocytes irrespective of the severity of the disease (3,9), and many studies have since shown a comparable response to COH in patients with varying degrees of endometriosis (2,3,6). Retrieval of a reduced number of oocytes secondary to technical difficulties encountered during the laparoscopic approach thus seemed the major factor contributing to a poor outcome associated with severe endometriosis in the earlier data. Furthermore, these studies may have included patients with reduced ovarian reserve as suggested by elevated basal FSH and E_2, which are known independent predictors of a poor outcome after IVF.

The study presented is singular in terms of the stringent criteria used for patient inclusion for data analysis. Not only was advanced age considered a factor influencing the outcome of IVF, but a much wider spectrum of variables was addressed including male-factor infertility and elevated day 3 FSH/E_2. Assisted hatching was also considered a criterion for exclusion, as it is suggested to improve implantation rates after IVF-ET in a subcategory of patients (13). Of the 17 patients excluded secondary to basal day 3 FSH levels greater than 20 IU/L, all were younger than 40 years of age; 9 of 17 had stage I/II, and 8 had stage III/IV endometriosis. The cause–effect relationship of this association of reduced ovarian reserve with early stages of the disease remains to be explored.

The significantly impaired fertilization rate of preovulatory oocytes retrieved from patients with moderate/severe disease in the study presented is of interest and consistent with observations in the literature (14,15). Wardle *et al.* (16) were the first to demonstrate an impaired fertilization potential of oocytes derived from patients with endometriosis compared with IVF of oocytes obtained from patients with tubal disease. Racowsky *et al.* (14) have demonstrated significantly impaired fertilization of preovulatory oocytes from patients with endometriosis as well as an increased incidence of aneuploidy with increasing severity of the disease, further suggesting an adverse impact of severe endometriosis on the biological behavior of oocytes. Given the exclusion of male-factor infertility from our patient group, this difference in fertilization rate of oocytes with severe stages of the disease suggests an intrinsic oocyte defect . Because comparable numbers of embryos were available for transfer in patients in the two groups and the outcome of IVF in terms of implantation and ongoing pregnancy rates was similar in patients with varying severity of the disease, IVF may compensate for the suboptimal behavior of oocytes in patients with severe endometriosis, thus contributing to an outcome comparable to that seen with lesser degrees of severity of the disease.

Some groups have demonstrated reduced pregnancy rates following IVF in patients with severe endometriosis (2–5). The exists limited data suggesting impaired implantation in patients with endometriosis, regardless of the severity of the disease (3,10). The mechanisms for implantation failure that have been hypothesized include an embryotoxic intrauterine environment and the presence of autoantibodies in a subcategory of patients with endometriosis (3,17), and more recently, aberrant integrin expression in the endometrium was found to be associated with endometriosis, suggesting a defect in uterine receptivity in some of these patients (18). Our results do not support these hypotheses, as the implantation rates of patients with varying stages of endometriosis were comparable, a finding concordant with that reported earlier by several groups (3,11,12). The difference in the ongoing pregnancy rates in our two groups of patients was 11%, a value not of statistical significance. For the observed difference in the ongoing pregnancy rate to be significant at the 20% level using a power of 80%, we would require the inclusion of a minimum of 93 patients in each arm of the study.

CONCLUSION

The data presented demonstrate that the outcome of IVF remains unaffected by increasing severity of the disease. There is a suggestion of an adverse impact of increasing severity of endometriosis on the biological behavior of oocytes, as exhibited by reduced fertilization rates of oocytes in patients with moderate/severe endometriosis. IVF may bypass the effects of this biological compromise, thus contributing to comparable outcomes with varying severities of endometriosis.

ACKNOWLEDGMENTS

The authors extend their appreciation to John T. Sawaya, M. D., and Zuying Chen, M. D., for their help with data compilation.

REFERENCES

1. American Fertility Society: Revised American Fertility Society Classification for Endometriosis. Fertil Steril 1985;43:351–352
2. Oehninger S, Acosta AA, Kreiner D, Muasher SJ, Jones HW, Rosenwaks Z: In vitro fertilization and embryo transfer (IVF/ET): An established and successful endometriosis. J In Vitro Fert Embryo Transfer 1988;5(5):249–256
3. Dmowski WP, Rana N, Michalowska J, Friberg J, Papierniak C, El-Roeiy A: The effect of endometriosis, its stage and activity, and of autoantibodies on in vitro fertilization and embryo transfer success rates. Fertil Steril 1995;63(3):555–562
4. Matson PL, Yovich JL: The treatment of infertility associated with endometriosis by in vitro fertilization. Fert Steril 1986;46(3):432–434
5. Inoue M, Kobayashi Y, Honda I, Awaji H, Fujii A: The impact of endometriosis on the reproductive outcome of infertile patients. Am J Obstet Gynecol 1992;167:278–282
6. Tummon IS, Colwell KA, MacKinnon CE, Nisker JA, Yuzpe AA: Abbreviated endometriosis associated infertility correlates with in vitro fertilization success. J In Vitro Fert Embryo Transfer 1991;8:149–153
7. Geber S, Paraschos T, Atkinson G, Margara R, Winston RML: Results of IVF in patients with endometriosis: the severity of the disease does not affect the outcome or the incidence of miscarriage. Hum Reprod 1995;10(6):1507–1511
8. Oliveness F, Feldberg D, Liu HC, Cohen J, Moy F, Rosenwaks Z: Endometriosis: A stage by stage analysis-the role of in vitro fertilization. Fertil Steril 1995;64(2):392–398
9. Arici A, Oral E, Bukulmez O, Duleba A, Olive DL, Jones EE: The effect of endometriosis on implantation: Results from the Yale University in vitro fertilization and embryo transfer program. Fertil Steril 1996;65(3):603–607
10. Simon C, Gutiérrez A, Vidal A, Santos MJ, Tarin JJ, Remohi J, Pellicer A: Outcome of patients with endometriosis in assisted reproduction: Results from in-vitro fertilization and oocyte donation. Hum Reprod 1994;9:725–729
11. Yovich JL, Matson PL: The influence of infertility etiology on the outcome of IVF-ET and GIFT treatments. Int J Fertil 1990;35(1):22–29
12. Chillik CF, Acosta AA, Garcia JE, Perera S, Van Uem JFHM, Rosenwaks Z, Jones HW: The role of in vitro fertilization in infertile patients with endometriosis. Fertil Steril 1985;44(1):56–61
13. Cohen J, Alikani M, Trowbridge J, Rozenwaks Z: Implantation enhancement by selective assisted hatching using zona drilling of human embryos with poor prognosis. Hum Reprod 1992;7:685–691
14. Racowsky C, Folkers MF, Klasener SO, Hutchinson-Cole H, Nelson-White T, Lewis A, Cui H, Gelety TJ: Abstracts of the 44th Annual Meeting of the Pacific Coast Fertility Society, April 17–21, 1996
15. Yovich JL, Yovich JM, Tuvik AI, Matson PL, Wilcox DL: In vitro fertilization for endometriosis. Lancet 1985;2:552
16. Wardle PG, McLaughlin EA, McDermott A, Mitchell JD, Ray BD, Hull MGR: Endometriosis and ovulatory disorder: Reduced fertilization in vitro compared with tubal and unexplained infertility. Lancet 1985;236–239
17. Weed JC, Arguembourg PC: Endometriosis: Can it produce an autoimmune response resulting in infertility? Clin Obstet Gynecol 1980;23:885–893
18. Lessey BA, Castlebaum AJ, Sawin SW, Buck CA, Schinnar R, Bilker W, Strom BL: Aberrant Integrin expression in the endometrium of women with endometriosis. J Clin Endocrinol Metab 1994;79:643–649