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Natural cycle IVF in unexplained, endometriosis-associated and tubal factor infertility

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BACKGROUND: To elucidate possible differences between unexplained and minimal peritoneal endometriosis-associated infertility, we studied their outcome in natural cycle IVF (NIVF). METHODS: A prospective cohort study was carried out on unexplained (33 couples), minimal peritoneal endometriosis-associated (30 couples) and tubal factor (24 couples) infertility in 223 NIVF cycles, using human chorionic gonadotrophin (HCG) for ovulation induction. RESULTS: During the first NIVF attempt, follicular and luteal phase oestradiol, FSH, LH and progesterone concentrations, as well as endometrial thickness and follicular diameter were similar among the three groups. Periovulatory follicular growth monitored from day of HCG administration to oocyte aspiration was significantly lowered in unexplained infertility compared with minimal endometriosis-associated and tubal factor infertility. The fertilization rate, clinical pregnancy rate per initiated cycle, per successful oocyte retrieval and per embryo transfer, in minimal endometriosis (80.0, 10.4, 16.0 and 23.5% respectively) were similar to that in tubal factor infertility patients (68.6, 5.8, 11.4 and 16.0%) but significantly higher ($P < 0.05$) than that of the unexplained infertility group (62.2, 2.6, 5.4 and 8.7%). CONCLUSIONS: The significant reduction in follicular periovulatory growth, fertilization and pregnancy rates in unexplained infertility compared with minimal peritoneal endometriosis patients may be explained by sub-optimal follicular development with possibly reduced oocyte quality, intrinsic embryo quality factors or by impaired implantation. From a clinical point of view, NIVF is less suited to unexplained infertility treatment, but might represent an interesting treatment option for minimal peritoneal endometriosis-associated infertility.

Key words: natural cycle/IVF/endometriosis/unexplained infertility/tubal factor

Introduction

Whether endometriosis is the cause or a consequence of subfertility is continuously debated, and the question of possible differences between unexplained and minimal peritoneal endometriosis-associated infertility remains unresolved (Audebert *et al*., 1992; Brosens, 1994; Crosignani and Vercellini, 1994; Koninckx, 1994; Evers, 1994, 1996, 2000). Its aetiology seems heterogeneous with possible explanations connected with endocrinology, immunology, genetics and reproductive physiology (Chan and Tucker, 1991; Ryan and Taylor, 1997; Pellicer *et al*., 1998). From a practical point of view, if unexplained and minimal peritoneal endometriosis-associated infertility respond identically to the same treatment, a possible difference in aetiology might be irrelevant. However, to administer optimal treatment, an obvious task would be to know exactly which mechanism to adjust and act accordingly. Furthermore, it would be in the interest of an infertile couple to receive individualized treatment, taking into consideration their own wishes based on the pregnancy rates for each treatment option, the cost and the possibility of adverse effects.

Study by Tanbo *et al*. comparing treatment outcome in conventional IVF, indicated that unexplained and minimal peritoneal endometriosis-associated infertility patients had similar outcomes (Tanbo *et al*., 1995). Significantly lower embryo cleavage rates in unexplained and minimal peritoneal endometriosis-associated infertility patients compared with tubal infertility (control) patients indicated gamete defects as possible causes of infertility. However, in other studies when patients with unexplained or minimal peritoneal endometriosis-associated infertility were treated with artificial insemination by the husband, a significantly higher pregnancy rate was achieved in the unexplained infertile patients compared with the endometriosis patients (Åbyholm *et al*., 1992; Omland *et al*., 1998), possibly reflecting different underlying causes in the development of these conditions.

In spite of improvements in assisted reproductive techniques, implantation rates are still low. Embryo quality, the interaction of the embryo and the endometrium as well as factors influencing implantation, such as the presence of pinopodes or adverse effects of gonadotrophins, appear to be of utmost importance (Paulson *et al*., 1990; Ertzeid *et al*., 1993; Simón *et al*., 1995; Giudice, 1999; Nikas, 1999). This prospective cohort study examining the outcome of IVF in a natural menstrual cycle

was initiated in order to examine possible differences in the causes of unexplained and endometriosis-associated infertility.

Materials and methods

Eighty-seven infertile couples, requesting assisted fertilization in our unit, were recruited for this prospective cohort study between 1992 and 2001 and were treated by natural cycle IVF (NIVF) prior to conventional IVF treatment (where necessary). Three different diagnostic infertility groups were studied: unexplained infertility (33 couples), minimal peritoneal endometriosis-associated infertility (30 couples) [American Fertility Society (AFS) stage 1] (American Fertility Society, 1985) and tubal factor infertility (24 couples). Inclusion criteria were: minimum 2 years of infertility, regular menstrual cycle (interval of 25–35 days) with luteal phase progesterone >15 nmol/l and normal prolactin, free thyroxin and thyroid stimulating hormone (TSH) concentrations. All couples had normal semen characteristics [World Health Organization (WHO), 1992] in two or more tests (i.e. $\geqslant 20\times10^6$ sperm/ml, ⩾50% of sperm with progressive motility, >30% normal morphology), female age <37 and male age <45 years old, were of good health and used no adjunct medication.

Diagnosis of the three groups was confirmed by laparoscopy or, in some cases of tubal infertility, by laparotomy prior to the initiation of the study. In the unexplained and endometriosis-associated infertility groups the laparoscopy was normal, except for the presence of minimal peritoneal endometriosis (AFS stage I) (American Fertility Society, 1985) in the latter. The tubal factor group was unselected containing patients with or without sactosalpinx, as our unit practised no systematic removal of tubes with sactosalpinx prior to infertility treatment during the study period. In the unexplained and endometriosis-associated infertility groups, all the females were primary infertile, while the tubal factor group was recruited among patients with occluded Fallopian tubes and primary or secondary infertility. The endometriosis-associated infertility patients had not received any medical or surgical treatment for endometriosis.

All patients were examined during a natural menstrual cycle by vaginal ultrasound scanning, measuring the diameter of the maturing follicles and the endometrial thickness. They were first seen in the early follicular phase (cycle days 2–4) in order to exclude cycles with ovarian cysts. Thereafter, ultrasound scanning was performed daily or every second day from cycle day 9 or 10, depending on cycle length, until the criteria for timing ovulation induction with HCG were met. These criteria were: follicle diameter of ⩾15 mm and oestradiol concentration >0.5 nmol/l and/or an endometrial thickness >7 mm. Normal follicular development was defined as daily expansion of at least 1–2 mm in follicle diameter prior to ovulation induction (Hackeloer *et al*., 1979). Serum hormone analysis was performed daily or every other day throughout the menstrual cycle. HCG was administered (5000 IU Profasi®; Serono, Aubonne, Switzerland) 34–36 h prior to follicle aspiration. The presence of an unruptured follicle was controlled shortly prior to follicle puncture. The retrieved oocyte was cultured and inseminated after standard IVF procedures (Åbyholm *et al*., 1990; Tanbo *et al*., 1995) and the resulting embryo was transferred to the uterus 48–72 h after oocyte retrieval. No luteal phase support was given. Fourteen days after oocyte retrieval, serum HCG was measured to confirm pregnancy. A biochemical pregnancy was defined by HCG >25 IU/l, while a clinical pregnancy was always verified by the presence of a vital fetus by vaginal ultrascan in week 5–6 after transfer.

If the attempt resulted in no pregnancy, the couples were offered a maximum of five consecutive NIVF cycles before treatment with conventional IVF. In these subsequent cycles, monitoring was performed by vaginal ultrasound scanning only to determine the optimal time for HCG administration, based on follicular size and endometrial appearance. In order to compare hormone values during the menstrual cycles, the day of HCG administration was designated day 0, the days prior to this day as follicular phase and the days following day 0 as luteal phase. Serum concentrations of oestradiol were measured using the Abbott IMx® Oestradiol assay (microparticle enzyme immunoassay, MEIA) (Abbott Laboratories, Abbott Park, IL, USA). Serum concentrations of progesterone were measured using Coat-A-Count radioimmunoassay (Diagnostic Products Corporation, Los Angeles, CA, USA) until November 1996 and by Access® Immunoassay System kit (Beckman Instruments, Chaska, MN, USA) from 1996 onward. During the period 1992–1996, serum concentrations of FSH and LH were measured using DELFIA (dissociation-enhanced lanthanide fluoroimmunoassay) kits (LKB Wallac, Turku, Finland), and from 1997, Access® Immunoassay System (Beckman Instruments, Chaska, MN, USA). Interassay coefficients of variation for the individual analyses were: oestradiol 6.0–20.0%; progesterone 5.5–9.0% (DPC) and 9.7–21.0% (ACCESS); FSH 3.6–3.8% (DELFIA) and 7.0–9.0% (ACCESS); LH 3.7–4.2% (DELFIA) and 10.0–14.0% (ACCESS). Normal serum ranges were as follows: oestradiol 0.11–2.10 nmol/l; progesterone <0.5–85.0 nmol/l; FSH <10 U/l; LH <12–75 U/l. Changing assay methods did not influence the range of the values.

Statistics

Continuous data are given as mean ± SD. Independent-sample *t*-test and one-way analysis of variance (ANOVA) were used to compare normally distributed continuous variables between two and three groups respectively. Non-parametric data were examined with the Mann–Whitney and Kruskal–Wallis tests. Comparison of categorical variables was performed with the χ^2 test or Fisher's exact test in the case of any infrequent response. Area under the curve (AUC) was calculated by the trapezoidal rule. P values < 0.05 were considered statistically significant.

Results

Patient characteristics are shown in Table I. No difference was found in age distribution or body mass index. The unexplained infertility group had significantly longer infertility duration before referral to our assisted reproductive unit and a longer time to NIVF after the diagnostic laparoscopy/laparotomy than either of the other two groups.

During the follicular and luteal phases of the first NIVF cycle, the three groups had similar values for oestradiol, progesterone, FSH and LH (area under the curve), as well as for cycle day 2/3 FSH and peak hormone concentrations on the day after HCG injection (Table II). Furthermore, there was no statistically significant difference between the groups in endometrial thickness or follicular diameter on the day prior to HCG injection, on HCG day, or on the day of follicular puncture. We observed a significant difference in follicular growth from HCG day to retrieval day in unexplained infertility (1.8 ± 1.0 mm) compared with endometriosis-associated (2.6 ± 0.9 mm, $P < 0.05$) and tubal factor infertility (3.3 ± 2.3 mm, $P < 0.05$). Normal follicular development was seen at least once in all but two patients with unexplained or tubal factor infertility.

As indicated in Table III, of 223 started NIVF treatment cycles 80 (35.9%) were cancelled prior to oocyte retrieval due to spontaneous ovulation (35.0%) or no follicular development

Table I. Patient characteristics

	Unexplained infertility	Endometriosis-associated infertility	Tubal factor infertility
Couples	33	30	24
Age (mean ± SD), years	30.8 ± 3.0	31.1 ± 4.0	31.1 ± 2.9
Body mass index (mean ± SD)	21.7 ± 2.7	23.2 ± 3.6	23.2 ± 3.6
Years of infertility (mean ± SD)	5.7 ± 2.7[a]	4.0 ± 2.1	4.2 ± 2.0
Years between laparoscopy/ laparotomy and NIVF (median, range)	1.1 (0.5–10.0)[a]	0.5 (0.2–4.0)	0.5 (0.1–5.0)

$P < 0.05$ versus both endometriosis-associated and tubal factor infertility groups.

Table II. Hormone concentrations, area under the curve (AUC) hormone concentrations, follicular diameter, and endometrial thickness during the first NIVF cycle. Day 0 is the HCG administration day

	Unexplained infertility	Endometriosis-associated infertility	Tubal factor infertility
Cycle day 2 or 3 FSH (IU/l)	6.0 ± 2.0	6.8 ± 2.1	6.7 ± 1.8
Follicular phase			
AUC oestradiol (day×nmol/l)	3.2 ± 1.0	3.3 ± 1.6	3.0 ± 0.9
AUC progesterone (day×nmol/l)	8.1 ± 2.0	5.9 ± 1.2	7.7 ± 1.8
AUC FSH (day×IU/l)	45.1 ± 15.8	45.2 ± 17.2	46.6 ± 18.2
AUC LH (day×IU/l)	45.8 ± 21.1	45.2 ± 19.0	58.2 ± 36.4
Day 1 after HCG injection			
oestradiol (nmol/l)	0.9 ± 0.3	1.0 ± 0.5	0.9 ± 0.3
FSH (IU/l)	5.3 ± 0.7	9.5 ± 6.4	6.4 ± 4.3
LH (IU/l)	20.4 ± 15.2	20.2 ± 14.4	33.0 ± 39.9
Luteal phase			
AUC oestradiol (day×nmol/l)	5.6 ± 2.5	6.5 ± 2.4	6.8 ± 2.7
AUC FSH (day×IU/l)	49.7 ± 25.3	53.2 ± 16.8	60.4 ± 18.5
AUC LH (day×IU/l)	60.3 ± 35.8	64.2 ± 31.3	68.4 ± 25.3
AUC progesterone (day×nmol/l)	261.1 ± 102.0	248.6 ± 91.7	230.8 ± 82.1
Follicular diameter			
Day –1 (mm)	15.9 ± 1.8	14.9 ± 1.7	14.8 ± 1.5
Day 0 (mm)	17.6 ± 1.7	16.8 ± 1.5	16.7 ± 1.2
Oocyte retrieval day (mm)	17.8 ± 3.8	19.8 ± 2.6	19.8 ± 2.5
Endometrial thickness			
Day –1 (mm)	7.8 ± 1.2	8.3 ± 1.6	7.0 ± 1.0
Day 0 (mm)	9.2 ± 1.2	8.3 ± 1.5	8.8 ± 1.3
Oocyte retrieval day (mm)	10.4 ± 2.0	9.6 ± 1.7	9.7 ± 1.4

Data are mean ± SD.

(0.9%). There were no statistically significant differences observed between the groups and from 143 retrievals, a total of 122 oocytes were collected. Ten unexplained infertility couples were unsuccessful in 21 NIVF attempts due to spontaneous ovulation (six couples), no follicular development (two couples) or no oocyte at retrieval (two couples). Five couples with endometriosis-associated infertility were unsuccessful in eight NIVF attempts due to spontaneous ovulation (three couples), no follicular development (one couple) or no oocytes at retrieval (one couple). Four couples with tubal factor infertility were unsuccessful in eight NIVF trials due to spontaneous ovulation (one couple), no follicular development (three couples) or no oocyte at retrieval (one couple).

The fertilization rate for the endometriosis-associated group was comparable with that of the tubal factor group, but was significantly higher than the unexplained infertility group ($P < 0.05$). No statistically significant differences were found in semen parameters among the group (Table III). Furthermore, the difference in repeated fertilization failure per couple was not significant: seven unexplained (7/37, 18.9%), two endometriosis-associated (2/50, 8.0%) and four tubal factor (4/35, 20.0%) infertile couples.

All fertilized oocytes in the unexplained and tubal factor infertility groups cleaved normally and were transferred on day 2/3 after oocyte retrieval. Among endometriosis-associated infertile women, six embryos were not transferred because of cleavage arrest. Per started NIVF cycle, the embryo transfer rates were not significantly different for unexplained (29.9%), endometriosis-associated (44.2%) and tubal factor (34.8%) infertility. The total number of embryos transferred, the total pregnancy rate per initiated cycle, per successful oocyte retrieval and per embryo transfer is given in Table III. The pregnancy rate per initiated cycle for endometriosis-associated infertility was similar to that of the tubal factor group but significantly higher than that of the unexplained infertility group ($P < 0.05$) (Table III). The pregnancy rate per successful

Table III. Results of NIVF cycles

	Unexplained infertility	Endometriosis-associated infertility	Tubal factor infertility	Total
Couples (*n*)	33	30	24	87
Started NIVF cycles (*n*)	77	77	69	223
Cancelled cycles prior to oocyte retrieval (*n*, %)	31 (41.6)	22 (28.6)	27 (39.1)	80 (35.9)
Oocyte retrieval attempts (*n*, %)	46 (59.7)	55 (71.2)	42 (60.9)	143 (64.1)
Collected oocytes (*n*) (%, oocyte retrieval)	37 (80.4)	50 (90.9)	35 (83.3)	122 (85.3)
Sperm samples (mean ± SD)				
Concentration ($\times 10^6$/ml)	109.7 ± 49.7	139.1 ± 79.6	98.2 ± 48.3	
Progressively motile (%)	67.2 ± 16.3	60.0 ± 17.7	58.7 ± 21.1	
Morphology (%)	35.2 ± 18.0	42.0 ± 11.7	42.7 ± 7.6	
Fertilized oocytes (*n*, %)	23 (62.2)[a]	40 (80.0)	24 (68.6)	87 (71.3)
PR/initiated cycle (%)	2.6 (2/77)[a]	10.4 (8/77)	5.8 (4/69)	6.3 (14/223)
PR/successful oocyte retrieval (%)	5.4 (2/37)[a]	16 (8/50)	11.4 (4/35)	11.5 (14/122)
PR/embryo transfer (%)	8.7 (2/23)[a]	23.5 (8/34)	16.0 (4/24)	17.1 (14/82)

[a]$P < 0.05$ compared with endometriosis-associated infertility.
PR = pregnancy rate.

oocyte retrieval was statistically lower ($P < 0.05$) for unexplained infertility compared with endometriosis-associated infertility, but was similar compared with tubal factor infertility (Table III).

Fourteen clinical pregnancies were obtained in total, with a significantly higher pregnancy rate per embryo transfer in the endometriosis-associated infertility patients than in unexplained infertility patients ($P < 0.05$) (Table III). Once again, there were no differences between the endometriosis-associated and tubal infertility groups. There were no biochemical pregnancies or spontaneous abortions and 14 healthy children were delivered.

Discussion

In this study we used NIVF as a tool to investigate possible aetiological differences between unexplained and endometriosis-associated infertility. NIVF gives an opportunity to explore pre-ovulatory follicular and endometrial growth. Successful cycles with embryo transfer reflect normal gametes with the ability to fertilize *in vitro*. In cycles where an oocyte is obtained but not fertilized, one or both gametes might be abnormal. Furthermore, in cases where an oocyte is not obtained or oocyte retrieval is difficult to time, the cause might be subtle variations in hormone secretion, making ovulation irregular. Since the adverse effect of controlled ovarian stimulation on the endometrium is omitted, a reduced implantation rate in NIVF cycles might reflect endometrial deficiency.

Normal follicular development as assessed by vaginal ultrasound scanning was observed in all but three couples during NIVF attempts; two unexplained infertility patients and one tubal factor patient. However, our finding of significantly lower follicular growth between day of HCG administration and day of retrieval in unexplained infertility compared with both minimal endometriosis-associated and tubal factor infertility, might indicate sub-optimal follicular development and reduced oocyte quality in unexplained infertility.

Recent investigations support a theory of altered folliculogenesis resulting in reduced quality oocytes as one of the causes of unexplained and endometriosis-associated infertility (Cahill *et al*., 1995; Harlow *et al*., 1996). Comparison of unexplained infertile with normal fertile controls has shown significant alterations in FSH and LH levels measured during the menstrual cycle, which might represent an early loss of ovarian reserve (Leach *et al*., 1997). The present study indicates no such differences between the study groups and the tubal factor control group. Compared with strict tubal factor infertility, unstimulated cycles from both minimal/mild endometriosis and unexplained infertility have been found to have significantly reduced serum LH in addition to reduced LH concentration in follicular fluid (Cahill *et al*., 1995). These findings could imply a dysfunction in the pituitary–ovarian function. A defective aromatase activity of granulosa cells has been suggested, as this activity and progesterone production were reduced in minor endometriosis-associated infertility compared with a control group of tubal factor and unexplained infertility (Harlow *et al*., 1996). This non-significant trend in unstimulated cycles became significant in stimulated cycles when more follicles were present. We found no such differences. However, the luteal phase in the present study was influenced by HCG ovulation induction, which may have masked any possible differences.

The empty follicle syndrome where follicle maturation and hormone levels seem normal both in unstimulated and stimulated cycles might occasionally contribute to infertility. Recurrence of the phenomenon is rare (Zreik *et al*., 2000) and was not seen in the present study. However, in one woman with minimal endometriosis, both empty follicles and atretic oocytes were observed during NIVF and no normal oocytes were obtained.

Defects in gamete function or interaction as causes of decreased fertilization and cleavage rates *in vitro* of both unexplained and minimal/mild endometriosis-associated infertility compared with tubal factor infertility have been demon-

strated in several publications (Åbyholm *et al*., 1990; Chan and Tucker, 1991; Fleming *et al*., 1994; Tanbo *et al*., 1995; Bergendal *et al*., 1998). Studies using donor gametes and IVF to differentiate between oocyte and/or sperm abnormalities have supported these findings. Donor sperm trials significantly increased fertilization rates in unexplained infertility couples but had no impact on fertilization rates in endometriosis-associated and tubal factor infertility couples (Hull *et al*., 1998). This finding might indicate an inability of fertilization in sperm characterized as 'normal' according to WHO criteria (World Health Organization, 1992). When comparing fertilization rates in NIVF, we found significant differences between the unexplained and endometriosis-associated infertility groups, supporting the presence of possible fertilization difficulties in the unexplained infertility group. The semen parameters were comparable between the groups.

Previous observations of implantation and pregnancy rates in endometriosis-associated infertility, after conventional IVF, are inconclusive as both reduced (Yovich *et al*., 1988; Simón *et al*., 1994, 1995; Arici *et al*., 1996) and comparable rates (Inoue *et al*., 1992; Mills *et al*., 1992; Dmowski *et al*., 1995; Geber *et al*., 1995; Tanbo *et al*., 1995) have been found compared with tubal factor infertility. However, in most of these studies, the endometriosis group consisted of all stages (AFS I–IV) (American Fertility Society, 1985). As donated oocytes from endometriotic ovaries had much lower implantation rates than tubal factor controls, and as oocytes from women without endometriosis donated to women with endometriosis showed the same implantation ability as tubal factor controls, more advanced stages of endometriosis could be responsible for poor oocyte quality (Simón *et al*., 1994). When performing controlled ovarian stimulation combined with artificial insemination with partner's spermatozoa, pregnancy rates for unexplained infertility were found to be superior to endometriosis-associated infertility, probably because peritoneal and consequently intra-tubal environments are more hostile when endometriosis is present (Åbyholm *et al*., 1992; Dmowski *et al*., 1994; Omland *et al*., 1998).

Implantation failure is an important limiting factor for the success of assisted reproduction techniques and probably also represents a contributing factor in subfertility. Hormones administered during controlled ovarian stimulation alter embryo–endometrial interaction in both humans and animals in spite of transfer of good quality embryos (Paulson *et al*., 1990; Ertzeid and Storeng, 2001). Biochemical markers of the implantation window have shown abnormal expression of integrins in women with luteal phase defects, unexplained infertility and minimal or mild endometriosis and might link their implantation failure to defects in uterine receptivity (Garcia-Velasco and Ariei, 1999). Furthermore, endometrial pinopode formation seems important for optimal implantation and their abnormal expression might compromise implantation in unexplained and endometriosis-associated infertility as in different assisted reproduction technology stimulation protocols (Nikas, 1999).

NIVF is the least invasive of IVF techniques and in spite of lower success rates, it remains a simple, low risk and low cost treatment option (Daya *et al*., 1995). Conventional IVF has significantly higher success rates per cycle, but also more adverse effects, such as multiple pregnancies, ovarian hyperstimulation syndrome and a possible negative long-term impact on the ovaries (Tarlatzis *et al*., 1995). For certain patients, the legal and ethical problems with surplus embryos make NIVF the treatment of choice. The cumulative live birth rate in NIVF cycles of 32% after four cycles, reported by Nargund *et al*., might further advocate the use of this treatment (Nargund *et al*., 2001). However, a major problem of NIVF is a relatively large cancellation rate, even when HCG is used for ovulation induction, due to premature LH surges and difficulties in timing oocyte retrieval. The administration of gonadotrophin-releasing hormone (GnRH) antagonist is promising in this respect, as cancellation rates seem to decrease significantly (Rongières-Bertrand *et al*., 1999). It would be interesting to observe whether NIVF cycles combined with GnRH antagonist and HCG could produce good oocytes and favourable conditions for implantation.

The results of this study indicate different aetiological mechanisms of unexplained and endometriosis-associated infertility. A hostile peritoneal environment could explain the lower success rate of endometriosis-associated infertility in artificial insemination with partner's sperm compared with both stimulated (Tanbo *et al*. 1995) and unstimulated IVF cycles. As pregnancy rates for unexplained infertility are comparable both in stimulated artificial insemination with partner's spermatozoa (Omland *et al*., 1998) and conventional IVF, but significantly decreased in NIVF, the actual increase in the number of fertilizable oocytes seems decisive. Hence, a frequently occurring gamete defect accounting for a relative inability to conceive in unexplained infertility might effectively be overcome when surplus gametes are provided.

The patients of this study were offered conventional treatment with IVF if pregnancy was not attained by NIVF. A follow-up study is projected.

In conclusion, the lower fertilization rate, pregnancy rates per initiated cycle, per successful oocyte retrieval and per embryo transfer in unexplained infertility patients compared with that of minimal endometriosis-associated infertility patients after NIVF treatment may be explained by either gamete or intrinsic embryo quality factors or by impaired implantation. From a clinical point of view, NIVF is less suited for unexplained infertility treatment, but might represent an interesting treatment option for minimal peritoneal endometriosis-associated infertility. The present finding of significantly lower follicular growth between day of HCG administration and follicular aspiration day in unexplained infertility patients compared with endometriosis-associated and tubal factor infertility patients, further indicates sub-optimal follicular development with possibly reduced oocyte quality.

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