Two-year Experience with Gamete Intrafallopian Transfer (GIFT) at Maharaj Nakorn Chiang Mai Hospital

Apichart Oranratnachai MD, Teraporn Vutyavanich MD,
Chamnong Uttavichai MD, Prayode Jongyusuk MD,
Pallop Pongsuthirak MD, Warunya Ittipunkul BSc,
Waraporn Piromlertamorn BSc.

Division of Reproductive Endocrinology and Infertility,
Department of Obstetrics and Gynaecology,
Faculty of Medicine, Chiang Mai University,
Chiang Mai 50002, Thailand

Abstract : *This article describes twenty patients who underwent gamete intrafallopain transfer (GIFT) at Maharaj Nakorn Chiang Mai Hospital during a two-year period. The details of this technique are described. The pregnancy rate, as defined by a rising of ß-hCG titer on two occasions at least 2 days apart, was 31.8% per treatment cycle, but the clinical pregnancy rate was only 27.4% per cycle. This technique could become an alternative to in vitro fertilization and embryo transfer (IVF & ET) in infertile women who have at least one patent fallopian tube. (Thai J Obstet Gynaecol 1991;3:95-102.)*

Key words : gamete intrafallopian transfer (GIFT), pregnancy rate, infertility

Recently many new reproductive techniques have been introduced with varying success rates to help infertile couples achieve pregnancies. These include superovulation with human menopausal gonadotropins (hMG)[1], intrauterine insemination (IUI)[2], intratubal insemination (ITI)[3], direct intraperitoneal insemination (DIPI)[4], peritoneal oocyte and sperm transfer (POST)[5], gamete intrafallopian transfer (GIFT)[6], zygote intrafallopian transfer (ZIFT)[7] and in vitro fertilization/embryo transfer (IVF/ET)[8]. In this report, we present our experience with the GIFT technique over a two-year period from October 1, 1989 to August 31, 1991.

Materials and Methods

Patient selection

All couples underwent complete basic infertility investigations, which included at least two semen

analyses, confirmation of ovulation with endometrial biopsy or mid-luteal serum porgesterone assays, post-coital tests and laparoscopic tubal patency tests. Patients with endometriosis were treated with expectant treatment and/or a course of danazol with or without surgery depending on the extent of the lesions. Those with cervical factor infertility were treated with intrauterine insemination with or without clomiphene citrate for ovulation induction. Oligospermic males, defined as a sperm count of less than 20 X 10^6/ml, were treated with mesterolone acetate for 4-6 months, with or without split ejaculate insemination. Couples with unexplained infertility were empirically treated with clomiphene citrate superovulation for 4-6 months. Prior to being considered for GIFT, each couple had exhausted all conventional treatment for their infertility problems as stated above. Only those with at least one patent fallopian tube were apporached for informed consent after detailed discussion of the GIFT procedure.

Controlled ovarian hyperstimulation

In our clinic, both long and short stimulation protocols, as previously described by others[9], are in use. The decision to place patients on either the long or the short protocol was made at the discretion of the attending physicians and not randomly. In brief buserelin acetate (Suprefact®, Hoechst) was administered intranasally at a dose of 100 μg, six times per day, starting in the mid-luteal phase of the preceding cycle in the long protocol, or on the first day of the current cycle in the short protocol. 2-4 ampules of human menopausal gonadotropins (hMG, Pergonal®, Serono) were given intramuscularly, according to the patient's age and previous response, from cycle day 3 onward. Pelvic sonogram was done on the first day of the cycle as baseline, and then on a daily basis from day 8 of the cycle to monitor follicular growth. Daily measurement of serum estradiol by radioimmunoassay was done on day 6 of the cycle. Human chorionic gonadotropin (hCG, Profasi®, Serono) 10000 IU was given intramuscularly when ultrasound demonstrated two or more ovarian follicles exceeding 17 mm in diameter an the estradiol level reached 300 pg/ml per dominant follicle. Buserelin was stopped on the day of hCG injection and oocyte retrieval via laparoscopy, minilaparotomy or vaginal aspiration under ultrasound guidance, was scheduled 34-36 hours later.

Sperm preparation

Male partners were requested to produce semen samples approximately 2.5 hours before oocyte retrieval. Semen was allowed to liquefy for 30-45 minutes before analysis and the results recorded on a standard form. Semen was gently mixed with Ham's F-10 (GIBCO, New York) at a ratio of 1:2 (vol/vol) and centrifuged for 10 minutes at 300 g. The superna-

tant was discarded and the sperm pellet resuspended in 1 ml of medium. After a second wash, 0.5 ml of medium was layered onto the loosened pellet and motile sperm were allowed to swim up for 45 minutes in a 5% CO_2 incubator at 37° C. Sperm concentration in the supernate was reassessed and adjusted to 100000/2.5 µl.

Oocyte preparation

Fluid from follicular puncture was examined carefully to recover oocytes under a dissecting microscope. Each harvested oocyte was placed in 1 ml of Ham's F-10 supplemented with 10% of the patient's serum in a 4-well plate, and kept in an incubator under 5% CO_2 in air at 37° C until transfer. After completion of oocyte retrieval, mature oocytes were selected and placed in a drop of Ham's F-10 supplemented with 50% of the patient's serum.

Tubal gamete transfer

Gamete transfer catheter (William A. Cook, Australia), with 1-ml tuberculin syringe attached at one end, was rinsed twice with Ham's F-10 supplemented with 50% of the paient's serum. Sperm and oocytes were loaded into the catheter in the following sequence: 10 µl of medium, an air space of 5 µl, 25 µl of sperm preparation containing 100000 sperm, an air space of 5 µl, one or more mature oocytes in 25 µl of medium, an air space of 5 µl and finally 10 µl of medium. The catheter was then introduced into the fimbriated end of the fallopian tube to a distance of 1.5-2 cm under laparoscopic control or under direct visualization in case of minilaparotomy. The content was gently expelled into the ampullary region of the tube. The catheter was then slowly withdrawn and returned to the laboratory in order to ensure that gametes were expelled. A similar procedure was repeated with the contralateral tube.

Luteal phase support

All patients received hCG (Pregnyl®, Organon) 1500 IU intramuscularly on the day of gamete transfer and on days 3, 6 and 9 after the transfer.

Diagnosis of pregnancy

Pregnancy was diagnosed on day 14 post-GIFT when the level of serum ß - hCG was greater than 25 mIU/ml, followed by a higher level in a subsequent assay 2 days later (Biochemical pregnancy). Clinical pregnancy was diagnosed when a gestational sac was visualized under vaginal ultrasound, which should be clearly seen from day 35 post-GIFT.

Results

Twenty-six infertile couples were enrolled for 28 cycles of GIFT. There were two patients who underwent the GIFT procedure twice. Of

these, six cycles in six patients were cancelled before oocyte retrieval; five because of poor ovarian response to hMG and one because of decreasing estradiol level before oocyte pick-up.

The average age of the twenty remaining patients was 34.9 years (ranged 29-42 years). All had fallopian tube patency on both sides. Fourteen of them had primary and six had secondary infertility, for an average duration of 7.8 years (ranged 2-14 years). Their infertility diagnoses are shown in Table 1. Two couples had both endometriosis and male factor infertility, one had both endometriosis and cervical factor infertility. Long and short ovulation induction protocols were used in equal number of cycles. On the average 25.8 ampoules of hMG were needed in the long protocol versus only 12.3 ampoules in the short protocol. However, there was no significant difference in the number of oocytes harvested. Pre-washed sperm motility increased from 43.1% to 77.2% after washing. An average of 4.7 oocytes were transferred per cycle (ranged 2-10). The gametes were transferred into both fallopian tubes in 10 cycles, resulting in 4 pregnancies. In 12 cycles, the gametes were deposited into only one tube, resulting in 3 pregnancies. The overall pregnancy rate was 31.8%, but clinical pregnancy rate was only 27.3% (Table 2).

Five pregnancies occurred in patients with endometriosis and one each in patients with cervical factor and unexplained infertility. There was no pregnancy in the six couples who had male factor infertility. The success of this procedure when stratified by the women's ages and by the number of oocytes transferred are shown in Tables 3 and 4.

Table 1 Infertility diagnosis

Diangnosis	Number
Endometriosis	13
Male factor	6
Carvical factor	2
Unexplained infertility	2

Table 2 Outcome of GIFT cycles

	Long protocol	Short protocol
Number of cycles	11	11
Number of hMG used (ampoules)	25.8	12.3
Number of oocytes harvested	5.3 (2-10)	4.7 (2-8)
Pregnancy	5	2
Biochemical	0	1
Clinical	5	1

Table 3 Pregnancy by women's ages

Age (years)	Number of patients	Number of pregnancy
<30	1	0
31-35	11	4
36-40	7	3
>40	1	0

Table 4 Pregnancy by the number of oocytes transferred

No. of oocytes transferred	No. of patients	No. of pregnancy
1-2	3	1
3-4	10	2
5-6	7	3
7 or more	2	1

Of the six clinical pregnancies there were two sets of twins, one of which aborted at 11 weeks pregnancy and the other is still on-going. Of the four remaining pregnancies, one aborted at 14 weeks, one was delivered preterm at 36 weeks (male fetus 2460 g), and two went on to term with uneventful deliveries.

The operating time for GIFT procedure was 74.5 minutes (ranged 46-95 minutes) when performed through minilaparotomy and only 25 minutes when done via laparoscopy. Accidental injury of small bowel occurred in one case during minilaparotomy, which was treated by primary closure, followed by an uneventful postoperative period. In this series there was one case with severe hyperstimulation syndrome, which responded very well to conservative treatment.

Discussion

The rationale for GIFT is that it allows gametes, i.e. sperm and oocytes, to be placed directly into the natural physiologic environment appropriate for fertilization. The introduction of spermatozoa within the ampulla of the tube may overcome compromised sperm transport in patients with male or cervical factor infertility[10]. Patients with unexplained infertility and endometriosis may have some impairment in the oocyte pick-up mechanism or in the transport of gametes to the normal site of fertilization or may have coexisting luteinized unruptured follicle syndrome[11-13], all of which could be bypassed by the GIFT procedure[6,10].

In our study, twice as many ampoules of hMG were needed in the long versus the short protocol, probably reflecting the absence of endogenous pituitary contribution when hMG stimulation is begun after down regulation has been accomplished[9]. However, it is still debatable whether one protocol is better than the other. Our data seems to support Mettler et al[14] who reported a higher pregnancy rate using the long protocol started in the mid-luteal phase compared with the short stimulation. On the other hand, Zorn et al[15] and Frydman et al[16] could not demonstrate any advantage of the long versus the short protocol regarding folliculogenesis, oocytes recovered and pregnancy rates. Until the advantage of either protocol is established, we feel that the choice should be tailored according to the patient's convenience, cost, and side-effects.

To date, no reliable biochemical testing has been developed which can accurately and rapidly provide direct correlation to oocyte matura-

tional status[17]. Only morphological criteria are available for grading of oocyte maturity into 4 or 5 aspects, based upon the degree of dispersion of the cumulus/corona cells and the presence of a polar body[18]. However, classification of the oocytes under the dissecting microscope is difficult, and to a large degree imprecise and subjective[17,18]. Therefore, in our laboratory we simply characterize the oocytes as mature or immature (types I-II vs III-IV)[19], and find the method to be simple and satisfactory.

Because of concern about multiple pregnancies of higher order (≥ triplets), many fertility centres restrict the number of oocytes transferred to 4 or 5[6,19,20]. However, in a large series of 1071 women, Craft et al[21] demonstrated a continuous increase in the overall pregnancy rates with the number of oocytes transferred i.e. 13.9% for 1-2 oocytes, 24.7% for 3-4 oocytes, 38.1% for 5-6 oocytes, 42.8% for 7-8 oocytes, 38% for 9-10 oocytes and 50% when more than 10 oocytes were transferred. In their series, multiple pregnancies were also increased, but to a far lower extent than expected, particularly in those patients who yielded more than 10 oocytes, and the number of oocytes replaced did not affect the abortion or the ectopic pregnancy rates. Given that the ideal number of oocytes that should be transferred is not known, we decided to replace all mature oocytes to avoid the problem of surplus eggs. On the average, 4.7 oocytes were replaced, resulting in an overall pregnancy rate of 31.7% which is comparable to that of Craft et al[21] and others[6,18-20]. So far, there are two sets of twins out of the six clinical pregnancies, with no ectopic pregnancy. However, we agree that in places where cryofacilities are available, it may be advisable to replace only 4 oocytes for each GIFT cycle, with consideration given to cryopreservation of excess eggs for transfer in subsequent cycles. The question of whether gametes should be transferred to only one or both fallopian tubes is still unsettled. Although we observed more pregnancies when oocytes were replaced into both tubes, the numbers are too small to allow any definite conclusion. Evidence to date suggests that unilateral GIFT may be at least as successful as the bilateral approach, while the operating time is shorter and there is less risk of trauma to the fallopian tubes at the time of gamete transfer[22].

Most GIFT/IVF centres use hCG or progesterone for luteal phase support in cycles stimulated with combined GnRH-a and hMG to counteract the luteolytic action of GnRH-a[9]. In our program we prefer to use hCG because there is one recent prospective double blind randomized trial demonstrating the advantage of hCG over progesterone[23]. However, in the case of ovarian hyperstimulation syndrome, progesterone is preferable in order to avoid further stimulation to the muliple corpora lutea[9].

Conclusion

Although the number of patients in this report is small, it confirms the notion that GIFT can be successfully applied to couples with a history of long-standing infertility, who have failed other conventional treatments. Compared with IVF, GIFT is less sophisticated and more physiologic in that it mimics the normal events involved in fertilization. We feel that GIFT is an attractive alternative to IVF in cases in which the female partners have at least one patent fallopian tube.

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