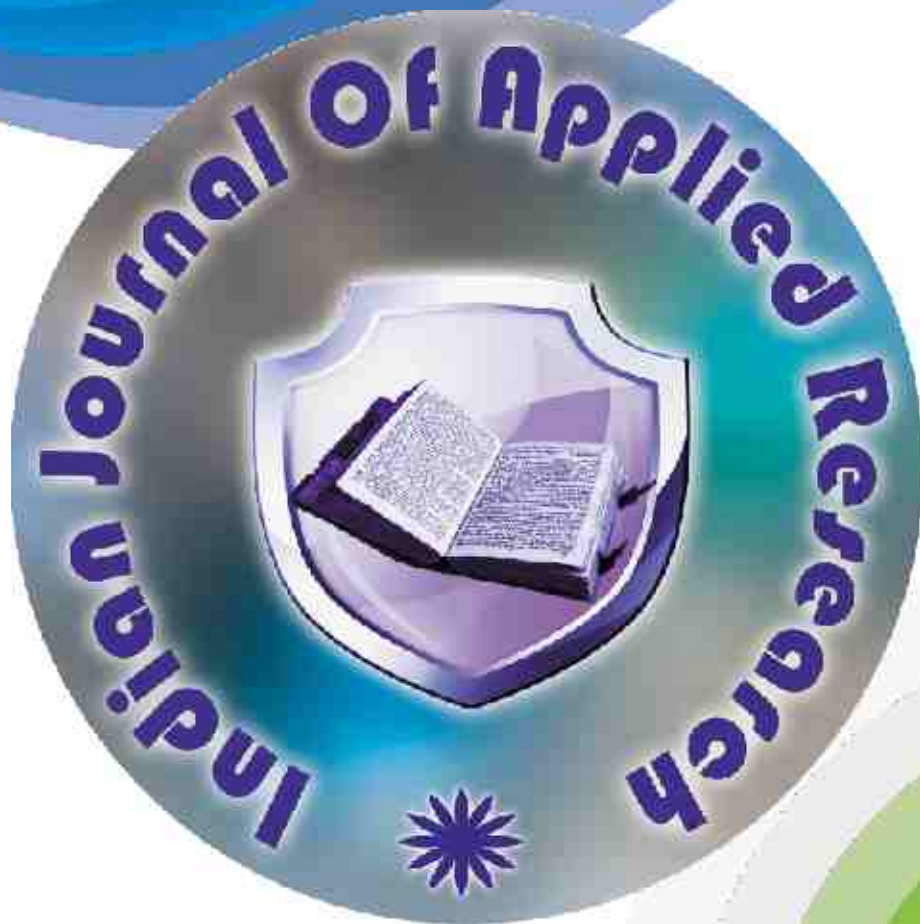


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## Research Paper

## Medical Science



## Advantage Of Fallopian Tube Sperm Perfusion Over Intra-uterine Insemination When Used In Combination With Ovarian Stimulation For The Treatment Of Unexplained Infertility.

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### ABSTRACT

200 consecutive patients with infertility in 404 stimulated cycles were included in the study. Those randomized to standard IUI included 100 patients in 184 cycles [158 clomiphene citrate/human menopausal gonadotrophin cycles and 26 Letrozole/FSH cycles exclusively for polycystic ovarian disease patients] (group A). Patients subjected to FSP included 100 patients in 220 cycles (193 clomiphene citrate/human menopausal gonadotrophin cycles and 27 Letrozole / FSH cycles exclusively for polycystic ovarian] (group B). Swim up semen preparation technique was used in all cases. Insemination was performed in both groups 34 hours after hCG administration. Standard IUI was performed using 0.5 ml of inseminate. In FSP 4ml inseminate was used. In group A (184 IUI cycles in 100 patients), 22 clinical pregnancies occurred (11.95% per cycle). In group B, (220 cycles of FSP in 100 patients), 48 clinical pregnancies occurred (21.81% per cycle). For non-tubal sub fertility, the results indicate clear benefit for FSP (Fallopian tube sperm perfusion) over IUI (Intrauterine insemination).

**Keywords :** Intrauterine insemination, Fallopian tube sperm perfusion.

### Introduction

Intrauterine insemination (IUI) with mild ovarian stimulation has been used for many years in the treatment of non tubal infertility. During IUI, pretreated semen is concentrated in a small volume of 0.5 ml and deposited by a catheter into the uterine cavity. The overall pregnancy rates reported in the literature ranged from 5.7% to 17.7% per cycle [1]. An alternative procedure, termed Fallopian tube sperm perfusion (FSP), has been reported with improved pregnancy rates in comparison with IUI [3, 4, and 5]. In FSP sperm preparation is identical to the used with IUI, but spermatozoa are diluted in a larger volume of medium up to 4 ml [3, 6]. This volume has been considered sufficient for bilateral passage of the spermatozoa through the fallopian tubes. Theoretically this would increase the density of capacitated spermatozoa near the oocytes and by consequence the higher pregnancy rates. A prospective randomized study was designed to determine whether FSP resulted in higher pregnancy rates than IUI.

### Material & Methods

Two hundred infertile patients, aged 17 to 39 years, undergoing 404 consecutive cycles of ovarian stimulation were studied from June 2008 to Jan 2010. Institutional board approval was obtained. These patients underwent a basic infertility workup. Exclusion criteria were age > 39 years, obstructed fallopian tubes, and cases with marked oligoasthenozoospermia (sperm count < 10X10<sup>6</sup> per ml). These patients underwent ovulation induction with clomiphene citrate and follicle stimulating hormone for non PCOS cases (170 patients) or Letrozole and FSH, for polycystic ovarian disease patients (70 patients). Induction protocol consist of clomiphene citrate 100 mg daily on days 3-7 of the cycle, and 75 IU daily of hMG (Human menopausal gonadotrophin) on days 6-9 of the cycle. In all PCOS patients, who had been on Metformin 500 mg t.i.d, Letrozole was given

orally in a dose of 2.5mg/day for 5 days starting from the third day of a spontaneous or progesterone induced menstrual bleeding. Inj purified FSH 75 IU administered on 6-9 day of cycle.

Cycles were monitored from day 9 transvaginal ultrasound measurement of the number and diameter of the growing follicles, the thickness and the morphology of the endometrium. A dose of 10,000 IU human chorionic gonadotrophin (HCG) was administered when at least one follicle had reached a diameter of 17 to 18 mm and at least 8 mm endometrial thickness with tri laminar 'halo' appearance seen. Patients were called 34 to 36 hour later, and either standard IUI (group A: 184 cycles in 100 patients) or FSP (group B: 220 cycles in the 100 patients) was performed. Maximum of four cycle treatment of IUI or FSP was considered for those patients who could not conceive in previous attempts. However those who failed to conceive with IUI were offered IUI only and vice versa. Semen samples were analyzed for density and motility using a fixed-depth counting chamber (Makler). The liquefied ejaculate transferred to a labeled sterile 14 ml round-bottomed disposable centrifuge tube (Falcon no.2095) and 4 ml flushing media (Medicult) added. After thorough mixing the sample centrifuged at 5000 rpm for 10 minutes. Then, the supernatants are discarded and pellet was resuspended and mixed in 3 ml of fresh flushing media (Medicult) and centrifuged for second wash again at 5000 rpm for 10 minutes. Each pellet is now gently layered with 0.5 ml for IUI and 4 ml for FSP of universal IVF media (Medicult), and incubated at 37°C in a humidified incubator with 5% Carbon dioxide for 1 hour. Intrauterine insemination was performed with conventional catheter using 0.5 ml of inseminate. In FSP 4ml inseminate was used and backflow of inseminate was occluded at cervical opening by allis clamp, which was suitably modified to avoid trauma to the cervix (Diagram-1) and was kept in place for about 3 to 4 minutes after insemination.



Values were recorded as mean  $\pm$  SD using Microsoft Excel version 4. Statistical analysis was performed using student's t-test for testing significance of difference between the means and the X2test to compute p-values for testing the agreement between observation and hypothesis (discrete data). The significance was defined as  $p < 0.05$ .

#### Results

The patient characteristics for group A and B were not significantly different concerning patient's age ( $28.42 \pm 2.78$  years and  $28.19 \pm 2.80$  years), type of sterility (primary infertility 74% versus 72% respectively), duration of infertility ( $5.6 \pm 2.1$  and  $5.3 \pm 1.9$  years respectively), and clinical indication for IUI or FSP (endometriosis 12% versus 12%, polycystic ovarian syndrome 34% versus 36%, cervical 4% versus 4%, unexplained 18% versus 12% and male factor sub fertility 32% versus 36%), ovarian stimulation (clomiphene citrate/hMG 85% versus 87% and Letrozole/FSH 15% versus 13%), number of follicles  $\geq 17$  mm diameter ( $3.93 \pm 1.37$  versus  $3.90 \pm 1.17$ ), day of hCG ( $12.8 \pm 3.4$  versus  $11.1 \pm 2.1$ ), endometrial thickness on the day of hCG ( $9.19 \pm 0.58$  mm versus  $9.14 \pm 2.1$  mm) and the number of spermatozoa ( $38.83 \pm 16.57 \times 10^6$  versus  $36.68 \pm 13.44 \times 10^6$ ) inseminated were not significantly different as shown in table-1 and 2.

Clinical pregnancy was defined by the presence of fetal heart beat, detected by ultrasound examination. Pregnancy rates were similar when compared for the etiology of infertility: for ovarian (PCOS) cause (17.7% versus 21.8%), endometriosis cause (8.4% versus 10.1%), for male infertility (12.8% versus 16.4%) and for unexplained infertility (14.4% versus 24%) for the two groups, respectively as shown in table-3. There was statistically significant difference in the overall pregnancy rate per cycle (11.95% per cycle for IUI versus 21.81% per cycle for FSP over four cycles) as shown in table-4. Two missed abortion occurred among the patients in group A (IUI) and one twin pregnancy. Three missed abortions occurred among the patients in group B (FSP) as well as two twin pregnancies. However, this limited number of abortions and multiple pregnancies are too low to allow testing for statistical significance. Three cases of mild ovarian hyper stimulation syndrome (OHSS) occurred in both groups.

#### Discussion

The purpose of this prospective, randomized study was to study pregnancy rates in couples with nontubal infertility when treated with FSP (inseminate volume 4 ml), in comparison with standard IUI (inseminate volume 0.5 ml). Pregnancy rates were 21.81 and 11.95% respectively. The same protocols for ovarian stimulation were used in both groups. There was no statistically significant difference regarding the age of the patients treated, the mean number of follicles, endometrial thickness on the day of hCG administration and the total number of motile spermatozoa inseminated.

Kahn et al. reported the first clinical experience with FSP [2]. In their study, they used a Frydman catheter for FSP and reported a pregnancy rate per cycle of 26.9% in patient with unexplained infertility and of 2.7% to 7.7% in patients with other etiologies. These excellent results, particularly in patients with unexplained infertility, were confirmed by other studies [1, 7]. Some investigators used a paediatric Foley catheter [5], a cervical clamp double-nut bivalve speculum [7]

and a very encouraging results were reported by Fanchin et al, in which FSP using an auto blocking device (FAST system) doubled their pregnancy rate from 20% to 40% [1]. The different types of catheters used for IUI have been compared but no study reported a significantly higher rate of pregnancy with any one of the catheters tested.

The FSP increases the intrauterine pressure, 70-200 mmHg, necessary for a flush influx of spermatozoa directly into the fallopian tubes, [1]. The high pregnancy rate per cycle for FSP as compared with standard IUI could be due to several causes as follows: first, the pressure injection of inseminate can either remove and/or circumvent transitory or partial obstruction of Fallopian tubes, such as that created by thick mucus or tubal polyps; second, the concentration of motile spermatozoa around the oocytes after FSP is higher than that obtained after standard IUI; and third, FSP leads to the inseminate overflowing into the pouch of Douglas. The more accepted hypothesis is the existence of a similar mechanical effect created following a hysterosalpingography, [10].

In this study, we tried to evaluate FSP not only in patients with unexplained infertility but also in patients with other causes of infertility including male causes. Two different stimulation regimes were used; however, the distribution of the two types of stimulation protocols (clomiphene citrate/hMG and Letrozole/FSH) appeared homogenous in both studies groups.

When comparing the pregnancy rates in both IUI and FSP in relation to the etiology of infertility, it is found to be statistically similar. However, there was statistically significant difference in the overall pregnancy rate per cycle (11.95% per cycle for IUI versus 21.81% per cycle for FSP over four cycles) as shown in table-4. Four studies [2, 4, 6, and 8] mentioned a maximum of three cycles per couple; one study [9] reported a maximum of four cycles. We also allowed maximum four cycle treatment of IUI or FSP before considering them for In vitro fertilization and embryo transfer (IVF-ET).

The type of catheter has no impact on the pregnancy rate after intrauterine insemination. We suitably modified the long size allis clamp, by attaching cervical occluding prongs with rubber cushions, and which was kept in place for about 3 to 4 minutes after insemination to prevent any significant reflux. Mild reflux does not seem to influence the results of the FSP but the significant reflux ( $> 0.4$  ml) would certainly reduce the pregnancy. If more than 1 ml comes back in the catheter, the operator need to wait for a few minutes and re-inseminate again. All of the authors agreed that the women tolerated the FSP technique very well. In our study some patients complained of post insemination pelvic transient pain, more so in FSP than in IUI. Other interesting domain of FSP application is the immunological infertility with presence of anti-spermatozoa antibodies. The existence of these antibodies does not always correlate with infertility.

From this study by comparing the overall results, we conclude that FSP, offers marginal advantage over the standard IUI, and could replace the IUI in certain indications for artificial insemination. However FSP is more expensive than IUI due to the increased media usages. It could be used as an alternative for couples with non tubal infertility before embarking on IVF treatment.



TABLE 1- Type of Infertility, clinical indications and ovarian stimulation protocols in group A (IUI) and B (FSP)

	Group A (Standard IUI)	Group B (FSP)	t-value	p-value
Type of Infertility				
Primary Infertility	74%	72%	0.450	0.652
Secondary Infertility	26%	28%	0.450	0.652
Clinical Indications for IUI/FSP				
Endometriosis	12%	12%	0.000	1.000
Ovulatory dysfunction	34%	36%	0.419	0.675
Cervical Factor	4%	4%	0.000	1.000
Male factor	32%	36%	0.844	0.399
Unexplained	18%	12%	1.695	0.090
Ovarian stimulation methods				
Clomiphene citrate/hMG (85%) (158 cycles in 86 patients)		(87%) (193 cycles in 88 patients)	0.579	0.563
Letrozole/FSH (15%) (26 cycles in 14 patients)		(13%) (27 cycles in 12 patients)	0.579	0.563

Group A : 100 patients,184 cycles Group B : 100 patients,220 cycles

Differences between groups A and B were not statistically significant as  $p > 0.05$

hCG = human chorionic gonadotrophin; IUI = intrauterine insemination, FSP = Fallopian tube sperm perfusion;

hMG = human menopausal gonadotrophin, FSH = follicle stimulating hormone;

TABLE 2- General characteristics, ovarian stimulation protocols, number of follicles, endometrial thickness on hCG administration and number of motile spermatozoa in groups A (IUI) and B (FSP)

	Group A (Standard IUI) (Mean $\pm$ SD) n=184 cycles	Group B (FSP) (Mean $\pm$ SD) n=220 cycles	t-value	p-value
Age (years)	28.42 $\pm$ 2.78	28.19 $\pm$ 2.80	0.825	0.409
Duration of Infertility (years)	5.6 $\pm$ 2.1	5.3 $\pm$ 1.9	1.506	0.132
No. of follicles $\geq$ 17mm diameter	3.93 $\pm$ 1.37	3.90 $\pm$ 1.17	0.237	0.812
Endometrial thickness (mm) on day of HCG	9.19 $\pm$ 0.58	9.14 $\pm$ 0.50	0.930	0.352
Number of sperm inseminated( $\times 10^6$ ) #	38.83 $\pm$ 16.57	36.68 $\pm$ 13.44	1.440	0.152
Day of hCG administration	12.8 $\pm$ 3.4	11.1 $\pm$ 2.1	6.146	0.000

Differences between groups A and B were not statistically significant except day of hCG administration

#Total number of spermatozoa with forward progressive motility

hCG = human chorionic gonadotrophin; IUI = intrauterine insemination, FSP = Fallopian tube sperm perfusion;

CC = clomiphene citrate; hMG = human menopausal gonadotrophin, FSH = follicle stimulating hormone;

TABLE 3 Clinical pregnancies (%) per cycle in relation to etiology of the infertility.

	Group A (Standard IUI) Patients 100 (Cycles =184)	Group B (FSP) Patients 100 (Cycles =220)	t-value	p-value
Mild/Moderate Endometriosis (CC+hMG protocol)	8.4% (12 patients, 24cycles)	10.1% (12 patients,27 cycles)	0.584	0.558
PCOS (Letrozol+FSH protocol)	17.7% (34 patients,61 cycles)	21.8% (36 patients,79 cycles)	1.027	0.304
Cervical causes (CC+hMG protocol)	0% (4 patients ,7 cycles)	0% (4 patients,9 cycles)	0.000	1.000
Unexplained (CC+hMG protocol)	14.4% (18 patients,33 cycles)	24% (12patients,26 cycles)	2.419	0.160
Male subfertility (CC+hMG protocol)	12.8% (32 patients, 59 cycles)	16.4% (36 patients ,79 cycles)	1.016	0.310

IUI = intrauterine insemination, FSP = Fallopian tube sperm perfusion; CC = clomiphene citrate;

hMG = human menopausal gonadotrophin, FSH = follicle stimulating hormone; PCOS= Polycystic ovarian syndrome,

TABLE 4- Clinical pregnancies (%) per cycles in relation to number of attempted treatment cycles in group A (IUI) and group B (FSP)

IUI				FS P			t - value	p - value
Treatment Cycle	No. of cycles	No. Of pregnancies	Pregnancy rate (%)	No. Of cycles	No. of pregnancies	Pregnancy rate (%)		
First	100	16	16.00	100	24	24.00	1.615	0.107
Sec ond	40	4	10.00	66	16	24.24	1.816	0.722
Third	30	2	6.66	34	06	17.64	1.326	0.189
Fourth	14	0	0	20	02	12.50	1.710	0.095
Total	184	22	11.95	220	48	21.81	2.608	0.009

IUI = intrauterine insemination, FSP = Fallopian tube sperm perfusion

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