Effect of Inseminated Volume on Intrauterine Insemination

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Submitted: January 3, 2001

Accepted: April 10, 2001

Purpose: Intrauterine insemination (IUI) is a method for the treatment of marital infertility involving the intrauterine or fallopian deposition of washed spermatozoa, depending on the amount of inseminated semen. In view of the divergent opinions about the inseminated volume, the objective of this study was to compare the two techniques (3.0 mL or 0.5 mL) in two groups of patients.

Methods: We performed 164 cycles of ovulation induction followed by IUI. The patients were divided into two groups according to the technique used. Group low volume – 50 cycles and 0.5 mL of inseminated semen; Group high volume – 114 cycles and 3.0 mL of inseminated semen. The cycle was monitored on the basis of endometrial thickness and follicular development measured by transvaginal ultrasound. Human chorionic gonadotrophin (hCG) was administered in the presence of a follicle measuring 18 mm in mean diameter. The procedure was performed after sperm washing using a discontinuous PureSperm[®] gradient, 40 h later.

Results: We obtained a similar clinical pregnancy rate for the two groups (14.0% for Group low volume and 15.7% for Group high volume). There was one abortion in each group. We detected no interference by any etiology of infertility or by the total motile recovered sperm with pregnancy rate.

Conclusions: The results did not demonstrate superiority of one method over the other, with both therapeutic alternatives being satisfactory for the treatment of infertile couples.

KEY WORDS: fallopian perfusion; infertility; intrauterine insemination; ovulation induction.

INTRODUCTION

The pregnancy rates obtained with intrauterine insemination (IUI) after ovarian hyperstimulation range from 4 to 40% (1–3) according to the technique used and the etiology of infertility. This variation is due to the fact that several studies have been conducted on limited populations using different protocols of ovarian stimulation, different groups of patients, and different insemination techniques (4–6).

Classical IUI implies the use of a semen volume of 0.5 mL or less with a high sperm concentration (7). In 1991, Kahn *et al.* (8) introduced a new method of IUI using a large semen volume (about 4 mL), which resulted in fallopian perfusion (FP). Using this technique, they were able to obtain better results in cases of sterility of no apparent cause (SNAC) in terms of pregnancy rate (25%) compared to those obtained by traditional IUI (8–10%).

Other studies have reported pregnancy rates higher than 25% with the use of FP depending on the etiology of infertility (2,9,10). However, when comparing the two techniques described in earlier paragraph, some authors did not detect an advantage of FP over classical IUI (6,7,11).

In view of these considerations, the objective of this study was to evaluate prospectively the results obtained by classical IUI and to compare them to those obtained by insemination of large semen volumes.

MATERIALS AND METHODS

Characterization of the Population Sample

We submitted 164 patients to 164 cycles of ovulation induction for IUI. All couples were seen at the Marital Infertility Outpatient Clinic of the Faculty of

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Medicine of Ribeirão Preto, University of São Paulo, from August 1999 to October 2000.

The investigation of infertility was conducted according to the protocol of the service, which includes measurement of follicle stimulating hormone (FSH), luteinizing hormone (LH), prolactin and thyroid stimulating hormone (TSH), transvaginal ultrasonography, hysterosalpingography (HSG), sperm count, sperm washing, and laparoscopy.

The couples were divided into two groups according to the IUI technique used. Group low volume consisted of 50 consecutive patients each submitted to one IUI cycle by the classical method with intrauterine infusion of 0.5 mL semen during the first 6 months of the study. Group high volume consisted of 114 consecutive patients each submitted to one IUI cycle with intrauterine infusion of 3.0 mL semen during the subsequent 9 months. The objective was to have two cases of infusion of a large semen volume for each case of classical infusion.

The inclusion criteria were that the patients should have a history of marital infertility of 2–6 years duration and the absence of use of hormonal contraceptives or of ovulation-inducing drugs during the 6 months preceding IUI. The exclusion criteria were that the women should be older than 40 years, total fallopian tube obstruction, and severe male factor (sperm concentration $<1 \times 10^6/\text{mL}$). After orientation and clarification of the procedures to be used, all couples signed a term of informed consent to participate in the study, which was approved by the Ethics Committee of the Institution.

Mean patient age was 31.7 ± 5.6 years for group low volume women and 31.1 ± 4.9 years for group high volume women. The distribution of the etiological factors of marital infertility for both groups is illustrated in Table I, with no significant difference between groups.

Table I. Cause^a of Infertility in the Low and High Volume Groups

	Group low volume (N = 50)	Group high volume (N = 114)	<i>p</i>
Male factor	15 (30.0)	52 (45.6)	ns ^b
Endometriosis	15 (30.0)	36 (31.5)	ns ^b
Ovulatory factor	9 (18.0)	39 (34.2)	ns ^b
Sterility of no apparent cause	7 (14.0)	10 (8.7)	ns ^b
Other factors	9 (18.0)	20 (17.5)	ns ^b

Note. Values represent *n* (%).

^a One patient can have more than one cause.

^b *p* > .05.

Procedures

Ovulation was induced by the administration of gonadotrophins (human menopausal gonadotrophin, hMG, or pure FSH, 75 IU/day intramuscularly) in combination with clomiphene citrate (CC), 50 mg, 1 tablet/day for 5 days starting between the 2nd and 5th day of the cycle, with the day when CC was first administered being Day 1 of induction. Gonadotrophins (1 ampoule/day at 4 P.M.) were administered on Days 2, 4, 6, and 8 of induction. In the presence of at least one follicle measuring 18 mm in mean diameter, 5000 IU of hCG were administered at 4 P.M. A new ultrasound examination was performed 40 h later to determine signs of ovulation and IUI was performed. IUI was not performed when more than three viable follicles were present or when no follicle reached 18 mm. Other reasons for discontinuation of treatment were personal problems. Fifty-six cycles were started during the formation of group low volume and 121 cycles were started during the formation of group high volume.

Semen was washed after liquefaction at room temperature and submitted to a discontinuous gradient of PureSperm[®] in a 1 mL column at 90%, in a 1 mL column at 45% plus 2 mL of semen and centrifuged at 1000 rpm for 30 min. The supernatant was then removed and 2 mL of HTF medium supplemented with synthetic serum was added to the pellet and centrifuged at 1000 rpm for 10 min. The supernatant was removed and resuspended in 0.5 mL (group low volume) or 3 mL (group high volume). The total amount of motile semen (forms A and B) was calculated.

Insemination was performed using a Kremer Delafontaine catheter (CCC Laboratory, Paris, France) inserted up to 0.5 cm from the uterine fundus. Group low volume received a rapid injection of 0.5 mL of the suspension and group high volume received a slow injection of 4 min duration of 3 mL using the same type of catheter. The cervix was pressed with a vaginal speculum in an attempt to prevent reflux, although sometimes this procedure was unsuccessful.

Statistical Analysis

Statistical analysis was performed using the Stata software and the tests applied were the *t* test for paired samples, proportion analysis, and the exact Fisher test for multiple variables. The results are presented as odds ratios (OR) and 95% confidence intervals (CI). The level of significance was set at *p* < .05 in all analyses. A power study was performed. On the basis of

Table II. Comparison of the Main Parameters Concerning Ovulation Induction in the Low and High Volume Groups

	Group low volume (0.5 mL)	Group high volume (3.0 mL)	<i>p</i>
Number of cycles	50	114	
Days of induction	11.4 ± 2.4	12.2 ± 2.9	ns ^a
Number of follicles (12–16 mm)	1.0 ± 0.9	1.2 ± 1.3	ns ^a
Number of follicles (>17 mm)	1.1 ± 0.5	1.4 ± 0.8	.02
Endometrial thickness on the day of hCG (mm)	10.4 ± 1.5	10.4 ± 2.3	ns ^a

^a *p* > .05.

previous experience at our center, a 15% pregnancy rate for IUI was expected, and to find twice the pregnancy rate, the CI set at 95% had a power of 60%.

RESULTS

Groups low and high volume were similar in terms of age, days of induction, number of follicles 12–16 mm in mean diameter, endometrial thickness on the day of hCG administration, and sperm concentration after washing. The number of follicles ≥17 mm in mean diameter was higher in group high volume (Table II).

The pregnancy rates were similar for the two groups studied, with 7 clinical pregnancies (14% of the inseminated cycles) in group low volume and 18 clinical pregnancies (15.7% of the inseminated cycles) in group high volume. No ectopic pregnancy occurred in either group. There was one abortion in each group. Table III presents the pregnancy rates according to total motile sperm inseminated in the two groups. The most satisfactory response in terms of the occurrence

Table III. Correlation Between Total Motile Retrieved Sperm and Pregnancy Rate for Group Low Volume (0.5 mL infusion, *N* = 50) and Group High Volume (3.0 mL infusion, *N* = 114) Patients

Total motile retrieved sperm	Pregnancy/cycle	
	Group low volume	Group high volume
<5 × 10 ⁶	1/9 (11.1)	2/12 (16.6)
>5 × 10 ⁶ < 10 × 10 ⁶	2/7 (28.6)	4/17 (23.5)
>10 × 10 ⁶	4/34 (11.7)	12/85 (14.1)
Number of pregnancies/ cycle	7/50 (14.0)	18/114 (15.7)

Note. Value in parentheses are percentages.

of pregnancy was observed in cases ranging from 5 × 10⁶ to 10 × 10⁶/total motile sperm inseminated.

The OR for predicting the success of high volume insemination was 1.29 (CI: 0.47–3.48). There was no relation between etiology of marital infertility and pregnancy rate (OR = 1.55, CI: 0.30–7.98 for male cause; OR = 1.30, CI: 0.23–7.32 for endometriosis; OR = 2.06, CI: 0.22–19.0 for SNAC). In groups low and high volume as a whole, pregnancy occurred in 15.7% of the cases of endometriosis, in 18% of the cases of male factor, in 20.8% of the cases of ovulatory factor, in 5.8% of the cases of SNAC, and in 20% of the fallopian tube factor of infertility.

DISCUSSION

The groups studied were quite homogeneous in terms of patient characteristics, with no significant difference in patient age or in total motile retrieved sperm for intrauterine insemination. Recent reports showed conflicting results when IUI techniques were compared, also in terms of volume of material deposited in the fundus region of the uterus during the procedure (2,7,12,13).

Evaluation of the results of different techniques on the basis of the etiology of infertility shows important divergences. Some studies (2,14) obtained significant results with IUI of 4 mL but the cases were not stratified according to infertility cause. Trout *et al.* (11), in a prospective randomized study and meta-analysis of the literature, detected high pregnancy rates (40%) only in patients with SNAC submitted to ovarian hyperstimulation and fallopian tube perfusion with high volumes of washed sperm, whereas other investigators (15,16) did not obtain success in these cases.

In the presence of other etiologies, the results of FP were similar to or even lower than those obtained by classical IUI (6). With respect to the male factor, Confino *et al.* (17) did not obtain pregnancy in a study with a mean number of three insemination cycles per patient, whereas Gerris *et al.* (18) reported excellent results using the same criteria. Studies evaluating the efficacy of IUI in cases of minimal or mild endometriosis have demonstrated a cumulative pregnancy rate of about 0.20 in 36 months. Ovarian stimulation is thought to favor the pregnancy rates, but the indication of this procedure is controversial (5,11,19). Our cases did not show better results according to etiology with either technique, although patients with SNAC corresponded to a small group.

When we compared the results of IUI with large volumes we detected no advantages. We recognize that the power of our analysis is low, but the clinical relevance of our number of cases is significant. In order to have an estimated difference between the two methods of at least twice the pregnancy rate, the number of cases should be approximately 1,000 patients, and the clinical impact of this finding has little relevancy. Our data confirm those reported by Schenk *et al.* (7) and Nuojuu-Huttunen *et al.* (6), who respectively obtained pregnancy rates per cycle of 7.9% and 8% with FP and of 21.7% and 20% with classical IUI. Although our patient series is smaller, we did not obtain better results with the infusion of large volumes in a group compounded by multiple etiologies. In contrast, Trout *et al.* (11) found better results with large infusion volumes only in patients with SNAC.

Indeed, Franco Júnior *et al.* (13) evaluated the volume of material deposited for IUI and the site involved by hysterosalpingography and noted that, starting from 0.4 mL, the contrast dye reached the uterus, isthmus, and tube ampulla, with no need for the infusion of large volumes to reach the fallopian tubes and a consequent lower cost for medium preparation.

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