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Slow release intrauterine insemination versus the bolus technique in the treatment of women with cervical mucus hostility

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Thirty-eight infertile women with cervical mucus hostility were divided at random into two groups for intrauterine insemination with prepared husband's semen. Eighteen women started with slow release (treatment A) and 20 with bolus (treatment B) intrauterine insemination in a cross-over study for four alternating cycles. Insemination was timed 30–36 h after a positive luteinizing hormone (LH) surge or injection of 5000 IU of human chorionic gonadotrophin, given at a follicular diameter of 18 mm during ultrasonically monitored, unstimulated cycles. A Grasby auto-syringe driver (type MS16) was used for the slow release intrauterine insemination to deliver 50 × 10^3 motile spermatozoa every minute for 3 h. Bolus intrauterine insemination was performed by deposition of 0.6 ml of prepared semen without changing the count from the swim-up portion of the washed spermatozoa. A total of 13 patients conceived, nine from 60 cycles of treatment A and four from 66 cycles of treatment B (chi-squared = 2.7143, $P < 0.05$ using one-tailed statistics).

Key words: insemination/intrauterine/slow release

Introduction

Intrauterine insemination (IUI) with husband's semen is widely used in the treatment of infertile couples with cervical mucus hostility and moderately abnormal male semen parameters. An important disadvantage of the bolus technique is the single deposition of a very large number of spermatozoa into the uterine cavity, in contrast to the prolonged slow release of small numbers of spermatozoa from the endocervix into the upper genital tract following natural intercourse. The procedure may have an adverse immunological impact (Beer and Neaves, 1978), may cause polyspermia (Englert *et al.*, 1986) and is a non-physiological procedure not incorporating the natural stages of sperm transport into the Fallopian tube. The entire length of the cervical canal, pre-treated with oestrogen, was estimated to contain 150 × 10^3, 180 × 10^3 and 53 × 10^3 spermatozoa 2, 24 and 48 h respectively following insemination (Insler *et al.*, 1981). By slow release, a persistent low concentration of spermatozoa is maintained at the oviduct in most mammals (Bishop, 1969), thereby prolonging the period of potential fertilization after a single coital act. Moreover, the practice of gamete intra-Fallopian transfer (GIFT) showed that only a few thousand motile spermatozoa are needed to achieve fertilization (Khan *et al.*, 1988). Accordingly, intrauterine insemination of low sperm numbers in a slow release pattern may be a more physiological process provided that sperm motility is adequate. During this study, we tried to simulate the natural cervical reservoir by using an external, slow release auto-syringe to inject small numbers of prepared motile spermatozoa in a continuous slow release pattern into the uterine cavity in patients with cervical mucus hostility. The research hypothesis, determined before treatment started, was directional (one-tailed), predicting a better response following slow release intrauterine insemination, compared to the bolus technique.

Materials and methods

Thirty-eight infertile women, 24–36 years old with primary or secondary infertility attributable to cervical mucus hostility, consented to participate in the study. Diagnosis was made by repeated negative post-coital tests (PCT) performed 8–12 h after sexual intercourse, with good cervical mucus biophysical characteristics (score ≥12) (Moghissi, 1977) and positive crossed hostility tests using Penetrak bovine mucus (Serono, Italy). All patients had regular ovulatory cycles as documented by repeated serum progesterone values of 33–72 nmol/l on day 21 of the cycle and normal pelvic findings as documented by hysterosalpingography and diagnostic laparoscopy. Patients with other infertility factors, including anti-sperm antibodies, were excluded from the trial. The husbands' semen analyses were repeatedly normal with a semen volume of 2.0–4.5 ml, sperm concentration of 26–150 × 10^6 per ml with ≥50% minimum progressive motility and ≥60% normal forms. Semen cultures for aerobic and anaerobic micro-organisms were negative. Serum anti-sperm antibodies were not detected in any patient using the gelatin agglutination test (GAT) according to the method described by Kibrick *et al.* (1952) and modified by Shulman (1978).

Patient allocation to the type of treatment in the first cycle was made at random by choosing an envelope containing the selection code. Eighteen patients started treatment with slow release intrauterine insemination (treatment A) and 20 with the bolus technique (treatment B) and were crossed-over in subsequent alternating cycles. Ovulation was monitored using an Aloka Echo

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Camera LS-Model SSD-248 ultrasonic machine with 3.5 MHz sector probe, utilizing the full bladder technique. This was supplemented by examination of the urine for luteinizing hormone (LH) surge using Quidel ovulation kits (La Jolla, CA, USA) starting on the ninth day of the cycle. Human chorionic gonadotrophin (HCG; Pregnyl, Organon, Holland) 5000 IU, was given 24 h after a follicle reached 18 mm without an LH surge, to guard against luteinization of unruptured follicles. Insemination was done 30–36 h after the HCG injection or LH surge. On the same day total leukocyte (WBC) and differential counts and erythrocyte sedimentation rate (ESR) were done. Semen was collected in sterile containers and an initial semen analysis was done immediately after sample liquefaction. Spermatozoa were washed in sterile Biggers, Whitter and Whittingham (BWW) medium containing 0.3% human serum albumin (Biggers *et al.*, 1971) using a standard two-step sequence (Wiltbank *et al.*, 1985). Spermatozoa were then overlaid with 1 ml of media and the swim-up portion of the specimen collected 30 min later.

For the slow release method of insemination, the sperm count was readjusted to 15×10^6/ml progressively motile spermatozoa in a 1 ml syringe attached to a 60 cm non-reactive plastic tube with a diameter of 1.5 mm. With the patient in the supine position, the cervix was exposed with a bivalve speculum, which was thoroughly cleaned with sterile gauze, and an endocervical swab was collected for aerobic and anaerobic culture. The plastic tube was introduced into the uterine cavity for 3 cm after being filled with the prepared spermatozoa and supported with vaginal sponges applied closely to the cervix. The speculum was then removed. The syringe was loaded on a continuous slow-release auto-syringe (type MS16, Grasby Medical, UK) adjusted to deliver 3×10^6 spermatozoa hourly (50×10^3 spermatozoa/min) for a total of 3 h. The auto-syringe driver was switched on and placed on the bed beside the patient.

Bolus intrauterine insemination was done by deposition of 0.6 ml of the sample prepared from the original swim-up portion of the washed spermatozoa, without changing the count. The rest of the swim-up portion was used for semen analysis.

Serum progesterone was measured using radioimmunoassay kits from RSL (Carson, CA, USA) 6 and 9 days after the HCG injection or LH surge. Endocervical swabs for aerobic and anaerobic culture, WBC and ESR were repeated 1 week after insemination. Treatment was offered for four alternating cycles unless pregnancy occurred. Patients were asked to keep a daily temperature chart record and to report any lower abdominal pain, vaginal discharge or any ill-health during the follow-up period.

Statistical analysis of the data was done using chi-squared test with one-tailed probability for the directional hypothesis (Clegg, 1990) and the Mann–Whitney test as applicable.

Results

There were no symptoms or signs suggestive of pelvic inflammatory disease during the follow-up period. The WBC and ESR values showed no significant changes after both types of treatment and none of the cervical swabs showed significant bacterial growth. Patients were treated for a total of 60 and 66 cycles with treatments A and B respectively. Eleven more cycles were abandoned because of failure to visualize a mature follicle. In the subsequent cycle, patients were offered the same treatment as for the abandoned cycle. All insemination cycles were ovulatory as documented by ultrasound monitoring and serum progesterone measurement during the luteal phase (≥ 30 nmol/l). One hundred and seven cycles (84.9%) had a spontaneous LH surge and HCG was given in 19 cycles (15.1%). There was no difference between the two groups regarding the distribution of these cycles. Thirteen pregnancies were achieved, nine after treatment A (15.0%) and four after treatment B (6.1%) (chi-squared = 2.7143, $P < 0.05$, using one-tailed probability). The cumulative pregnancy rate was 63.1% and 22% after treatments A and B respectively. Three pregnancies occurred in cycles supplemented with HCG (15.8%) whereas 10 occurred in natural cycles (9.3%) (chi-squared = 0.72403, $P > 0.05$).

There was no difference in semen parameters between the partners of women who conceived and those who did not ($P > 0.05$). The mean (95% confidence interval) sperm count, motility and proportion of abnormal forms of samples from partners of patients who conceived was 81.8×10^6 spermatozoa/ml (range $63.6-100 \times 10^6$), 66.9% (61.9–71.9) and 30.4% (26.6–34.2) respectively. The corresponding values in partners of patients who did not become pregnant were 76.4×10^6 spermatozoa/ml ($64.0-88.8 \times 10^6$), 67.4% (65.2–69.8) and 27.4% (24.5–30.3) respectively. The total number of spermatozoa inseminated during treatment A was 9×10^6/cycle. The corresponding mean value in treatment B cycles (95% confidence interval) was 31.5×10^6 spermatozoa/cycle ($30.1-33.0 \times 10^6$) ($P < 0.001$).

Discussion

This small randomized cross-over study showed that the pregnancy rate was three times higher after slow release intrauterine insemination compared to the bolus technique in patients with cervical mucus hostility. We previously reported five pregnancies following pulsatile intrauterine insemination in 20 women who failed to conceive after six treatment cycles with the bolus technique (Muharib and Abdel Gadir, 1989). Patients were initially apprehensive towards the use of intrauterine catheters but there was a shift of preference during the period of the study towards the 'automated' method of treatment, which might reflect a favourable placebo effect. The disadvantage of restricted patients' activity could be lessened by using self-retaining intrauterine catheters. This will allow prolongation of the insemination time and patients' ambulation after strapping the pump to the patient's thigh. Particularly relevant in this context is that the prolonged use of intrauterine catheters was not associated with any clinical evidence of induced pelvic inflammatory disease. Moreover, the use of such catheters may stimulate local prostaglandin production, which may improve the transport of spermatozoa along the Fallopian tube.

It can be concluded that the pregnancy rate may be improved by using slow release intrauterine insemination with a small number of prepared spermatozoa compared to the bolus technique, in the treatment of patients with cervical mucus hostility. Accordingly, it is essential to establish the optimal duration and number of spermatozoa required for continuous slow release intrauterine insemination, to obtain the maximum

pregnancy rate. This information may help in the treatment of patients with reduced sperm motility or count. Further work is planned in this area.

References

Beer,A.E. and Neaves,W. (1978) Antigenic status of semen from the view points of the female and male. *Fertil. Steril.*, **29**, 3–22.

Biggers,J.D., Whitten,W.K. and Whittingham,D.W. (1971) The culture of mouse embryos in vitro. In Daniel,J.C. (ed.), *Methods of Mammalian Embryology*. Freeman, San Francisco, 36 pp.

Bishop,D.W. (1969) Sperm physiology in relation to the oviduct. In Hafez,E.S.E. and Blandau,R.J. (eds), *The Mammalian Oviduct*. University of Chicago Press, 231 pp.

Clegg,F. (1990) Hypotheses. In Clegg,F. (ed.), *Simple Statistics, a Course Book for the Social Sciences*. Cambridge University Press, Cambridge, 57 pp.

Englert,Y., Puissant,F., Camus,M., Degueldre,M. and Leroy,F. (1986) Factors leading to tripronucleate eggs during human in-vitro fertilization. *Hum. Reprod.*, **1**, 117–119.

Insler,V., Glezerman,M., Bernstein,D., Zeidel,I. and Misgav,N. (1981) Cervical crypts and their role in storing spermatozoa. In Insler,V., Betendorf,G. and Geissler,K.H. (eds), *Advances of Diagnosis and Treatment of Infertility*. Elsevier, North Holland, New York, 195 pp.

Khan,I., Camus,M., Staessen,C., Wisanto,A., Devroey,P. and Van Steirteghem,A.C. (1988) Success rate in gamete intra-fallopian transfer using low and high concentrations of washed spermatozoa. *Fertil. Steril.*, **50**, 922–927.

Kibrick,S., Belding,D.L. and Merrill,B. (1952) Methods for the detection of antibodies against mammalian spermatozoa (II): a gelatin agglutination test. *Fertil. Steril.*, **3**, 430–436.

Moghissi,K.S. (1977) Significance and prognostic value of postcoital test. In Insler,V. and Bettendorf,G. (eds), *The Uterine Cervix in Reproduction*. George Thieme, Stuttgart, 231 pp.

Muharib,N. and Abdel Gadir,A. (1989) Pulsatile intrauterine insemination: a new and more physiological approach. The International Federation of Fertility Societies World Congress, Marrakesh.

Shulman,J.F. (1978) Agglutinating and immobilizing antibodies to spermatozoa. In J.Cohen and W.F.Hendry (eds), *Spermatozoa, Antibodies and Infertility*. Blackwell Scientific Publications, Oxford, 91 pp.

Wiltbank,M.C., Kosasa,S. and Rogers,B. (1985) Treatment of infertile patients, by intrauterine insemination of washed spermatozoa. *Andrologia*, **17**, 22–30.

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