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Evaluation of Vaginal Misoprostol Effect on Pregnancy Rate after Intrauterine Insemination

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Abstract: The goal of this study was to evaluate the effect of misoprostol on pregnancy rate after intrauterine insemination. This randomized double blind clinical trial study was performed on 66 (33 cases and 33 controls) infertile women who referred to infertility center of Imam Khomeini Hospital Sari, Iran for intrauterine insemination during 2006-2007. The two groups were matched for age, infertility causes and BMI. After intrauterine insemination, 200 mcg misoprostol was placed in posterior fornix of case group and a similar placebo tablet in control group. Chemical and clinical pregnancies and complications were recorded. Results were analyzed by means of SPSS 11 software, paired t-test and student t-test. The p-values of less than 0.05 were considered to be statistically significant. Chemical pregnancy (positive BHCG) occurred in 6 patients (18.2%) in each group. Clinical pregnancy occurred in 5 patients (15.15%) in case and 6 patients (18.2%) in controls. There were no significant statistical differences in complications between the two groups. Vaginal misoprostol after intrauterine insemination does not improve pregnancy rate.

Key words: Intrauterine insemination, misoprostol, prostaglandin, vaginal

INTRODUCTION

Infertility means having no pregnancy in sexually active couples after one year with no contraception (Berek, 2007). The prevalence is estimated 14% and the most important causes are: sperm dysfunction, disorders of ovulation, salpingitis, endometriosis, endometrial dysfunction and undetermined causes. There are various treatments to resolve the problem of infertility, such as ovulation induction with oral and injectable drugs, intrauterine insemination (IUI) IVF, ICSI sperm donation, egg donation and surgery, that are used according to the related reason of infertility (Gibbs *et al*., 2008). If the patient does not respond to ovarian stimulation, the next step will be IUI, which is cheaper and less invasive in comparison to other procedures (Yavas and Selab, 2004).

IUI is done by intrauterine injection of washed sperms in the cases of male factor, cervical, immunological and unexplained causes (Berek, 2007; Centola and Ginsburg, 2004). The success rate of IUI cycles depends on the fertility reason. For example, pregnancy rate of IUI in the cases of male factor is approximately 4.8% and in the cases of undetermined factor is about 11.6% (Berek, 2007). If IUI is not successful, other ART procedures will be used which are expensive and not desirable to most patients (Zeyneloglu *et al*., 2002).

According to earlier studies it seems that prostaglandins enhancing fertility via increase myometrial contractility and isthmic tubal relaxation affect luteal maintenance, immuno suppression and enhance spermatozoon-oocyte binding (Ni *et al*., 2003; Brown *et al*., 2001; Aref and Hafez, 1976). A few researches have been done in these cases. On the other hand, the studies have been controversial results regarding the effect of prostaglandins on success rate of IUI (Zeyneloglu *et al*., 2002; Ni *et al*., 2003; Brown *et al*., 2001; Aref and Hafez, 1976; Barroso *et al*., 2001; Cong *et al*., 2006; Kelly *et al*., 1991; Lu *et al*., 2001; Conte *et al*., 1985).

The aim of this study was to determine the rate of fertility after IUI and misoprostol use which has been done in Imam Khomeini Education and Research Hospital, Sari, Iran.

MATERIALS AND METHODS

This study is a randomized and double blinded clinical trial on 66 infertile patients, 33 patients and 33 as control. They were candidates for IUI and had been referred to infertility center of Imam Khomeini Hospital Sari, Iran, during 2006-2007.

All patients had been given written consent for the procedure. In the first visit of couples, their medical

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histories were reviewed, physical examinations were performed and their menstrual cycles and ovulation algorithm were studied. Laboratory tests including thyroid function tests and serum prolactin level, liver function tests and kidney function tests were requested. Hysterosalpingogram or laparoscopy for determining fallopian tubes, uterus and also semen analysis were done and causes of infertility were determined. The patients, whose infertility were due to PCOS, mild male factors or unexplained, were chosen for IUI. All patients' information including age, weight, height and BMI were recorded in the questionnaire. The procedure was fully explained to the patients and they gave their written consent.

All study protocols were approved by medical ethical committee of Mazandaran medical university.

The patients were asked not to have intercourse during 72 h before and after IUI and not to use antihistamine and NSAIDS.

The patients who met entry criteria were divided and matched in to 2 groups, based on infertility reason (PCOD, mild male factor, unexplained infertility) , BMI (19-25,26-30,31-35) and age (20-25y/o-26-30y/o-31-35y/o).

The first day of their period was established as first day of cycle. On the third day of cycle, transvaginal sonography was done for evaluation of ovaries and if the size of follicles was less than 5 mm, they were prescribed 100 mg Clomiphene Citrate daily (50 mg tab of Darupakhsh, Iran) for one week.

From the fourth day of cycle one amp of HMG (75 IU Darupakhsh, Iran) was given IM every other day. From the tenth day of cycle the growth of the follicles was evaluated with transvaginal sonography and if needed HMG were continued daily, until the dominant follicle reached 17 mm.

At this time, for induction of ovums, HCG 10000 IU was given IM and the number(s) of follicles which were more than 17 mm were recorded.

36-38 h after HCG injection, transvaginal sonography for assessing ovulation was done and IUI was performed on the same day. Sonography was done by two medical doctors and with the sonography Honda HS2000 model. If the ovulation was not confirmed, the patient was omitted from the study. From one day after IUI, 50 mg of progesterone was given IM daily (Abureihan company, Iran), to help the reinforcement of luteal phase. Sixteen days after IUI, BHCG was checked and if positive ($\geq$25 MIU mL^{-1}), chemical pregnancy was considered and the result was recorded. In these patients transvaginal sonography for evaluation of pregnancy was done four weeks after IUI and if there was evidence of fetal heart activity the clinical pregnancy was recorded. The sperm samples for IUI were collected in our clinic and were prepared by using standard method SUIM UP (Centola and Zavos, 1991). IUI was done by standard method (Ransom *et al*., 2000) by using Pierre Charron catheter. After remove of catheter from the cervix, a 200 μg tab of misoprostol (TFizer company, Italy) was inserted in the posterior fornix of the vagina in case group. In control group the placebo was inserted by the same manner. The patients were followed for 1 h and then were discharged.

The patients were informed about the probable complications and requested to report the following symptoms: diarrhea abdominal pain, nausea, vomiting and bleeding. All the complications were recorded. Placebo tablets were prepared in Pharmaceutical College of Mazandaran Medical Sciences and were similar to Misoprostol tablets. Neither doctors nor the patients knew the placebo from the drug. The data were analyzed with usage of SPSS (11) software. The two groups were compared for statistical calculation, with paired t-test, student t-test and χ^2. $p<0.05$ was considered significant.

RESULTS

Sixty six patients were enrolled in this study; 33 patients in misoprostol group and 33 patients in control group. They were matched and divided according to age, BMI and infertility causes.

The average age of case patients were (26.27±4.9) years and in control group (26.13±4.3) years ($p = 0.91$) (Table 1). The mean BMI were 26.20±3.1 kg m^{-2} in case group and in the control group 26.03±3.3 kg ($p = 0.83$)(Table 2). The mean level of serum FSH was 5.98±3.5 mIU mL^{-1} in case group and 6.85±4.1 mIU mL^{-1} in the control group ($p = 0.49$) and the mean level of serum LH was 7.1±5.3 mIU mL^{-1} in case group and 6.5±5.3 mIU mL^{-1} in control group ($p = 0.73$).

Table 1: Patient's variable data among groups

Variables	Case group (n = 33)	Control group (n = 33)
Causes of infertility		
PCO	5 (16.7%)	5 (16.7%)
Male factors	10 (33.3%)	10 (33.3%)
PCO+male factor	7 (21.2%)	7 (21.2%)
Unknown	11 (33.3%)	11 (33.3%)
Age		
20-25	15 (45.5%)	15 (45.5%)
26-30	13 (39.9%)	13 (39.9%)
31-35	5 (16.7%)	5 (16.7%)
BMI		
19-25	16 (48.5%)	16 (48.5%)
26-30	17 (51.5%)	17 (51.5%)

Table 2: Patient's demographic data

Variables	Case group	Control group	p-value
Age	26.27±4.3	26.13±4.3	0.91
Height (cm)	161.17±5.9	157.23±5.7	0.14
Weight (kg)	68.03±8.4	64.53±9.8	0.12
BMI	26.20±3.1	26.03±3.3	0.83
Total	30.00	30.00	

Table 3: Sperm data of counts variables among two groups

Variables	Case group (n = 33)	Control group (n = 33)	p-value
Sperm count before IUI million mL^{-1}	73.76±21.7	79.18±23.9	0.36
Sperm morphology before IUI %	57.83±8.6	56.07±11.5	0.51
Sperm motility % before IUI	55.33±9.8	56.43±10.1	0.67
Sperm count after IUI million mL^{-1}	42.20±22.3	46.71±20.7	0.42
Morphology after IUI %	70.69±16.5	76.26±17.5	0.22
Sperm motility after IUI %	84.70±12.1	85.64±15.9	0.80

There were no significant statistical differences in any of the above data. There were 6 patients with positive BHCG in each group (18.2%). There were 5 clinical pregnancy in the case group (15.5%) and 6 in the control group (18.2%). This difference was not significance statistically. The causes of infertility in both groups were similar and included one case of PCOD, two cases of male factor, one case of PCOD plus male factor and one case of unexplained infertility in misoprostol group there was one more PCOD in control group.

There were no significant statistical differences in the mean number of sperm count in seminal fluid, normal motility rate and normal sperms morphology before and after processing (Table 3).

In the follow-up of patients in the case group after usage of misoprostol abdominal pain and cramps occurred in 12 cases (36.4%) and in control group 9 cases (27.3%) were found (p = 0.66). Spotting was seen in 8 of the case group (24.24%) and 6 of the control group (p = 0.16). There were no significant statistical differences in these complications.

In 2 patients (6.06%) after IUI and misoprostol administration severe vaginal bleeding plus significant abdominal cramps were found. These symptoms were resolved spontaneously after 6-12 h. There was no chemical pregnancy in these two patients. We had no similar cases in the control group (p = 0.75). There was one patient in the case group who had diarrhea (3.33%), it resolved spontaneously (p = 0.51).

DISCUSSION

The role of decreased level of prostaglandin such as PGE in etiology of unexplained infertility was mentioned first in 1947 (Asplund, 1947). Similar studies on laboratory animals and human were done after that (Brown *et al*., 2001). Natural prostaglandins are divided in to 9 main groups (A-I) and all of them are unsaturated, hydroxylated fatty acids that are produced from arachidonic acid and resulted from the effect of cyclo-oxygenase on arachidonic acid (Breyer *et al*., 2000; Lim *et al*., 1999; Tamioka *et al*., 2000; Jakobsson *et al*., 1999; Murakami *et al*., 2000). Seminal fluid contains large amounts of prostaglandins (Kennedy *et al*., 2003; Templeton *et al*., 1978; Bendvold *et al*., 1984).

PGF, PGE and 19-hydroxylated PG are the most important type of them (Bendvold *et al*., 1987). The most active form of seminal fluid prostaglandin is PGE, that its range is variable from 2 to 272 mcg mL^{-1} of seminal fluid (Templeton *et al*., 1978). Although there is some evidence of decreasing prostaglandin concentration in seminal fluid of infertile couples with unknown etiology, the physiological effect of the decreasing infertility have not been cleared (Bygdeman *et al*., 1970).

Because of complication occur following intrauterine injection of seminal fluid, only washed sperm is injected in to uterus. Therefore, the supportive effects of PGs are limited (Brown *et al*., 2001). Misoprostol is a synthetic analog of PGE1 that in comparison with its natural form has stronger, longer lasting and more selective effect if used orally. In its pure form it is lucent oil. Misoprostol is marketed as oral tablets of 100 and 200 mcg (Katzung, 2004). This drug is usually used as protective agent in peptic ulcers following long term usage of NSAIDS. It is also used in induced of abortion, cervical ripening and labor induction (Brown *et al*., 2001). It is probable that its usage after IUI has similar effect as seminal fluid PGS during coitus, causing increase in fertility rate (Brown *et al*., 2001; Barroso *et al*., 2001; Cong *et al*., 2006). Based on earlier studies, it seems that PGS especially PGE may helps fertility rate, through facilitation of sperm transfer by relaxing cervical isthmus, increase uterine and fallopian tubes contractions, facilitating sperm penetration in the ovum and helping in suppressing female immune response to spermatozoa (Ni *et al*., 2003; Brown *et al*., 2001; Aref and Hafez, 1976).

In current study use of misoprostol following IUI did not show any significant statistical difference in pregnancy rate comparing to placebo. Barroso *et al*. (2001) in a similar study showed that there was a higher rate of pregnancy in patients who received vaginal misoprostol. This study was not double blinded and they did not matched for BMI and infertility causes. In kipping with us Billiet *et al*. (2008) and Zeyneloglu *et al*. (2002) in their study concluded that there was no difference between IUI plus misoprostol with IUI alone. Brown *et al*. (2001) showed a higher pregnancy rate with IUI plus misoprostol. They used multiple medications and the highest pregnancy rate was in patients who received Clomiphen alone or in natural cycles had IUI. The pregnancy rate was the same in patients who received Clomiphen and FSH, similar to this study. In Brown's study there was more pregnancy in the patients who

received only FSH for induction, but it was not statistically significant. This findings with the result of current study raises the question weather gonadotrophins especially combined with clomiphen, causes a decrease in possible favorable effects of misoprostol in increasing fertility rate? and if it does, what is the process? In this study there was heavy bleeding and abdominal cramps in two patients who had received misoprostol. These complications occurred in less than an hour after IUI and pregnancy test also were negative in these two patients. Although Zeyneloglu *et al.* (2002) used just 50 μg of misoprostol, but due to occurrence of bleeding and abdominal cramps in 55% of patients, he had to stop his study and it is interesting that in Browns study (2001) in spite of 400 mcg misoprostol both case and control groups did not have any difference in the rate of pain and vaginal bleeding. In their study 2% of patients who received misoprostol had severe abdominal pain (1%) but no vaginal bleeding was seen. This noticeable difference was because of the form of drug usage. In order to make the drug similar to the placebo, Brown mixed misoprostol with triglyceride base. Whether triglyceride plus misoprostol have caused decrease complications due to sustained release of drug or other pathophysiologic conditions in the patients cause the difference? It needs further studies.

Therefore until benefits of this medication have not been proven or finding ways to decrease the side effect of the medication, we do not recommend the use of misoprostol in IUI cycles.

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