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ARTICLE

# Effects of treatment of ectopic pregnancy with methotrexate or salpingectomy in the subsequent IVF cycle

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**Abstract** Ectopic pregnancy is a known risk for patients treated with IVF. The objective of this study was to evaluate the effect of methotrexate (MTX) and laparoscopic salpingectomy as treatments of ectopic pregnancy on ovarian response during IVF cycles. Data of all women treated for ectopic pregnancy as a result of IVF treatment were evaluated; the study included women who had an unruptured ectopic pregnancy after IVF treatment that was treated with either MTX or laparoscopic salpingectomy and underwent a subsequent IVF cycle. The main outcome measures were baseline serum FSH concentrations and ovarian response in the subsequent IVF cycle after treatment of ectopic pregnancy. Of a total of 58 patients, 36 were previously treated with MTX and 22 others by salpingectomy. No significant differences were observed between the MTX and the salpingectomy groups in the parameters of ovarian response in the subsequent IVF cycle. Repeat ectopic pregnancy was encountered in one patient in each group with a total rate of 3.4% (2/58). No significant differences were found in the outcomes of the subsequent pregnancy after treatment with MTX or salpingectomy. It is concluded that neither prior MTX treatment nor salpingectomy affect ovarian response in the subsequent IVF cycle.



**KEYWORDS:** ectopic pregnancy, IVF, methotrexate, ovarian response, salpingectomy

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## Introduction

Ectopic pregnancy is a known risk for patients treated with IVF (Chang and Suh, 2010). The incidence of ectopic pregnancy among the IVF population varies from 1% to 8.6% (Chang and Suh, 2010). The apparent increase in the incidence of ectopic pregnancy after IVF could be due to the inherent baseline characteristics of the infertile women, such as tubal factors and previous surgery for endometriosis (Malak et al., 2011), which are known risk factors for ectopic pregnancy. Also, high steroid hormone concentrations associated with IVF treatment might impair tubal function. In addition, inadvertent embryo transfer into the Fallopian tube or transfer of high number of embryos might play a role (Chang and Suh, 2010; Oriol et al., 2008). Ectopic pregnancy is still the leading cause of pregnancy-related death in the first trimester (Khan et al., 2006) and immediate surgery is the only treatment option for haemodynamically unstable patients with a ruptured ectopic pregnancy (Vichnin, 2008). Appropriate treatment options for selected patients include expectant management when the β-human chorionic gonadotrophin (HCG) concentration is low and declining, medical treatment with methotrexate (MTX) and surgical treatment with salpingostomy or salpingectomy (Farquhar, 2005; Vichnin, 2008).

MTX is a folic acid antagonist and works by competitively binding to the enzyme dihydrofolate reductase which is required for DNA and RNA synthesis (Barnhart et al., 2007). It affects rapidly proliferating cells such as trophoblasts and has been used as a medical treatment for ectopic pregnancy. However, MTX can affect the proliferating germinal cells in the ovary. Reduced ovarian reserve after MTX treatment has been reported (Oriol et al., 2008). Salpingectomy can also affect the ovarian function by impairing the ovarian blood supply and reducing antral follicle count (Chan et al., 2003).

There have been a few studies evaluating the effect of MTX (Vichnin, 2008; Orvieto et al., 2007; McLaren et al., 2009) and salpingectomy on the ovarian reserve (Almog et al., 2011) (Xi et al., 2012). Yet, a study comparing the effects of MTX and salpingectomy on ovarian reserve has never been reported. The objective of this study was to evaluate the effects of MTX or salpingectomy as treatments of ectopic pregnancy on ovarian response in the subsequent IVF cycle.

## Materials and methods

This study evaluated data of all women treated for ectopic pregnancy as a result of IVF treatment from seven academic reproductive centres in Canada and Israel. The study was approved by the Institutional Review Board (reference number 11-689-SDR, granted 10 January 2012). The inclusion criteria were the following: (i) an unruptured ectopic pregnancy after IVF treatment that was treated with either MTX or laparoscopic salpingectomy; (ii) the patients underwent a subsequent IVF cycle within 12 months after the ectopic pregnancy; and (iii) the IVF treatment took place between the years 2005–2012. The study excluded patients whose ectopic pregnancy was treated with conservative management and those who had had frozen embryo transfers.

The combined database contained baseline characteristics of the patients including age, body mass index, cause and duration of infertility and baseline (day 3) concentrations of oestradiol, FSH and LH. Unfortunately, some of the participating centres did not have information about antral follicle count or anti-Müllerian hormone. Information about the ectopic pregnancy included gestational age at diagnosis, size of the pregnancy sac (the average of two diameters of the pregnancy sac), presence/absence of fetal heart beat, serum β-HCG concentration at treatment and type of treatment for the ectopic pregnancy. Additional data were the parameters of the IVF cycle resulting in ectopic pregnancy and of the subsequent IVF cycle. These included endometrial thickness, number of eggs that were collected and fertilized and number of embryos that were transferred and their quality. The embryos were graded according to the following criteria: grade 1, evenly sized blastomeres; grade 2, no more that 10% fragmentation; grade 3, fragmentation of no more than 50%; and grade 4, fragmentations of greater than 50%.

A sample size for this retrospective study was calculated with a power of 80% to detect a 20% difference in the number of oocytes collected between the two groups with two-sided alpha levels of 0.05. Using sample size calculation for unpaired two-sample t-test, a sample size of over 150 participants in each group was estimated. This estimation is based on the mean number of oocytes collected of 10.2 with a standard deviation of 6.6 (Almog et al., 2011).

### Statistical analysis

Shapiro-Wilk test was used to evaluate the distribution of the data. Comparisons were analysed using Student's t-test or Mann–Whitney *U*-test when appropriate and the results were presented as mean and standard deviation or median and interquartile range (IQR). Proportions were compared with chi-squared test or Fisher's Exact test. *P*-value $<0.05$ was considered significant.

## Results

Of a total of 58 patients, 36 were previously treated with MTX and 22 others by laparoscopic salpingectomy. None of the patients in the laparoscopy group received MTX. In the MTX groups, one ectopic pregnancy was diagnosed with fetal heart beat. Three pregnancies were not located by ultrasound and diagnosed as extrauterine of unknown location. Except these three, all others were tubal pregnancies. In the salpingectomy group, all ectopic pregnancy were located by ultrasound and were tubal. Three pregnancies were diagnosed with fetal heart beat. Each patient was treated with the same ovulation induction protocol before and after ectopic pregnancy. **Table 1** shows the baseline characteristics of the patients and the ectopic pregnancy. Apart from the serum β-HCG concentration just before the treatment of ectopic pregnancy, other parameters were comparable. The serum β-HCG concentration at treatment in the salpingectomy group (median 1730 (IQR 657–3861 IU/l)) were higher than those in the MTX group

**Table 1** Characteristics of women treated for ectopic pregnancy with methotrexate (MTX) or salpingectomy and who subsequently underwent another IVF cycle.

| | *MTX (n = 36)* | *Salpingectomy (n = 22)* |
|---|---|---|
| Age (years) | 33.8 ± 5.2 | 32.9 ± 4.8 |
| Body mass index (kg/m$^2$) | 26.2 ± 5.6 | 24.0 ± 5.8 |
| Basal FSH (IU/l) | 6.6 ± 1.8 | 6.9 ± 1.7 |
| Duration of infertility (months) | 36 (24—36) | 36 (24—60) |
| β-HCG concentration at treatment (IU/l)[a] | 547 (140—951) | 1730 (657—3861) |
| Gestational age at treatment (weeks) | 6.0 ± 0.66 | 6.2 ± 0.7 |
| Size of the pregnancy sac (mm) | 20.7 ± 6.1 | 23.3 ± 12.3 |
| Time between ectopic pregnancy and next IVF cycle (months) | 7.4 ± 6.0 | 5.8 ± 4.9 |
| Aetiology of infertility | | |
| Tubal | 6/36 (16.7) | 7/22 (31.8) |
| Unexplained | 13/36 (36.1) | 7/22 (31.8) |
| Ovulation | 1/36 (2.8) | 0/22 (0) |
| Endometriosis | 0/36 (0) | 1/22 (4.5) |
| Male factor | 16/36 (44.4) | 7/22 (31.8) |

Values are mean ± SD, median (interquartile range) or *n*/total (%).
[a]*P* = 0.04.

(median 547, (IQR 140—951 IU/l), 95% CI 16—2850 IU/l; *P* = 0.04). The average size of the pregnancy sac was also larger, but not significantly in the salpingectomy group (23.3 ± 12.3 mm) compared with the MTX group (20.7 ± 6.1 mm). **Table 2** demonstrates that baseline serum FSH concentration as an index of ovarian reserve was not affected by prior MTX or surgical treatment for ectopic pregnancy. Antral follicle count was only available for some of the patients (25 in the MTX group and 14 in the salpingectomy group) and was lower in both groups after treatment, although not significantly. When ovarian responses during the IVF cycle were compared before and after treatment,

**Table 2** Comparable IVF parameters before and after treatment for ectopic pregnancy.

| | *MTX (n = 36)* | | *Salpingectomy (n = 22)* | |
|---|---|---|---|---|
| | *Before* | *After* | *Before* | *After* |
| Basal FSH (IU/ml) | 6.6 ± 1.8 | 7.2 ± 2.5 | 6.9 ± 1.7 | 6.2 ± 1.5 |
| Antral follicle count[a] | 9.6 ± 5.1 | 6.5 ± 2.1 | 22.5 ± 4.9 | 17.2 ± 12.1 |
| No. of stimulation days | 10.6 ± 1.9 | 10.5 ± 1.4 | 9.1 ± 2.7 | 9.4 ± 2.8 |
| Total FSH dose (IU) | 3025 ± 1953 | 3021 ± 1868 | 2813 ± 1071 | 2895 ± 979 |
| Endometrial thickness at HCG day (mm) | 9.9 ± 2.0 | 9.6 ± 2.7 | 9.7 ± 1.7 | 9.8 ± 2.0 |
| Peak oestradiol concentration (pmol/ml) | 3645 ± 3156 | 3665 ± 2364 | 3887 ± 2379 | 3907 ± 2281 |
| No. of eggs collected | 10.2 ± 5.4 | 9.5 ± 5.3 | 13.4 ± 9.5 | 11.1 ± 7.6 |
| Fertilization rate | 57.4 | 63.5 | 56.4 | 61.7 |
| No. of embryos transferred | 2.7 ± 1.2 | 2.6 ± 1.1 | 2.6 ± 0.9 | 2.4 ± 0.9 |
| Embryo quality score | 1.9 ± 0.7 | 1.8 ± 0.7 | 1.9 ± 0.7 | 2.2 ± 0.8 |
| Pregnancy outcome | | | | |
| Ectopic pregnancy | 36/36 (100) | 1/36 (2.8) | 22/22 (100) | 1/22 (4.5) |
| Missed abortion | — | 1/36 (2.8) | — | 2/22 (9.1) |
| No pregnancy | — | 27/36 (75.0) | — | 15/22 (68.2) |
| Biochemical pregnancy | — | 0 | — | 3/22 (13.6) |
| Ongoing pregnancy | — | 6/36 (16.7) | — | 1/22 (4.5) |
| Cancellation (no response) | — | 1/36 (2.8) | — | 0 |

Values are mean ± SD, % or *n*/total (%). No statistically significant differences were found in the cycle characteristics when comparing pre- and post-treatment cycles in either the MTX or salpingectomy group. Also, no statistically significant differences were found in the subsequent cycle outcomes between the MTX and salpingectomy groups.
[a]AFC was available for 25 in the MTX group and 14 in the salpingectomy group.

there were no significant differences in either the MTX or the surgical group (**Table 2**). Repeat ectopic pregnancy was encountered in one patient in each group (total rate 3.4%, 2/58). No significant differences were found in the outcomes of the subsequent pregnancy after treatment between MTX and salpingectomy groups (**Table 2**).

## Discussion

Infertile patients, especially those who have experienced an ectopic pregnancy after IVF treatment, are different from women in the general population. They are treated with IVF after a period of infertility and wish to undergo another treatment as soon as possible after an episode of a failed IVF pregnancy. Therefore, there is a need to identify a treatment of ectopic pregnancy that has no effect on the ovarian reserve and the subsequent ovarian response to IVF treatment.

A few studies have evaluated the effects of MTX treatment of ectopic pregnancy and subsequent ovarian reserve and ovarian response to IVF treatment. Orvieto et al. (2007) showed that MTX treatment for an ectopic pregnancy did not influence the subsequent ovarian response to IVF treatment. Another study (Oriol et al., 2008) demonstrated that MTX treatment of ectopic pregnancy did not influence serum anti-Müllerian hormone concentrations in subsequent cycles. On the other hand, McLaren et al. (2009) concluded that there was a time-limited decreased oocyte yield during ovarian stimulation in women previously treated with MTX for ectopic pregnancy. In their study, when an IVF cycle occurred within 180 days of MTX exposure, a significant decline in retrieved oocytes was observed. The current findings support the previous two studies demonstrating no effect of MTX on subsequent ovarian response. The interval between the MTX treatment and the next IVF cycle was 7.4 ± 0.6 months. Whether a shorter interval will affect the ovarian response to an IVF cycle remains to be seen.

The effect of salpingectomy on the ovarian response was evaluated by four studies. Shulman et al. (2002) found that adnexal surgery is not detrimental to ovarian function. Almog et al. (2011) found that salpingectomy for hydrosalpinx did not influence the ovarian response during ovarian stimulation. Xi et al. (2012) studied the effect of salpingectomy for ectopic pregnancy among IVF patients and found no differences in the IVF parameters before and after salpingectomy treatment. On the other hand, Orvieto et al. (2011) observed a significant decrease in the ipsilateral ovarian response following salpingectomy, as reflected by the quantity of developing follicles during ovarian stimulation for IVF. The current findings are in accordance with the first three studies demonstrating that previous salpingectomy does not influence ovarian response to IVF treatment. However, it is important to excise the hydrosalpinx close to the tube to avoid compromising the blood supply to the ovary.

The weakness of this study is its retrospective nature. The lack of standardization and randomization is reflected in the differences of β-HCG concentration and the size of the ectopic pregnancy between the groups: a more advanced pregnancy would get a salpingectomy rather than MTX.

Despite gathering data from seven IVF units with a total of 6000 IVF cycles annually, only 58 cases could be included, which is far fewer than the 150 participants per arm required by the sample size calculation. This can be partially explained by the fact that many cases of ectopic pregnancies or subsequent treatment cycles that resulted from frozen embryo transfer cycles had to be excluded because the effect of treatment on ovarian response could not be compared. Despite this limitation, this study provides assurance that both MTX and salpingectomy can be safely used in the treatment of an ectopic pregnancy in the context of IVF. An interesting finding was the lower clinical pregnancy rates in both groups in the subsequent IVF cycle: 19.4% in the MTX group and 13.6% in the salpingectomy group. Some studies evaluated the pregnancy rate after ectopic pregnancy but in the general population not in IVF patients. One study (Oriol et al., 2008) evaluated the pregnancy rate in IVF patients after MTX treatment and found a pregnancy rate of 33.4% after treatment; however, their study included only 14 patients. In the current study, lower pregnancy rates were found despite apparently similar ovarian responses, numbers of oocytes retrieved and embryos transferred and quality of the embryos in both groups. The only parameters that seemed to be affected by the treatment were the antral follicle count and number of collected eggs (especially in the salpingectomy group). In both groups these parameters were lower after treatment, although the differences were not statistically significant, probably due to the low number of patients. It could be that with a larger sample size significant differences would be seen, which could explain the lower pregnancy rate. Folliculogenesis is a process thought to require almost an entire year in the human ovary (Gougeon, 1986). The majority of this time (270 days) is spent in the gonadotrophin-independent phase (preantral phase), while the remaining 85 days are gonadotrophin dependent. As the follicle transitions from the quiescent primordial state, it gains not only gonadotrophin receptors but also an increased blood supply (McLaren et al., 2009). It is possible that MTX and salpingectomy (due to reduced blood supply) affect those follicles as seen in the reduced antral follicle count. Another effect could be on oocyte quality or endometrial receptivity. However, this current observation of low pregnancy rate after ectopic pregnancy in IVF patients is preliminary and should be evaluated in further larger studies.

Certainly each type of treatment has its indications and criteria including serum HCG concentration for MTX treatment. Here, patients in the salpingectomy group had higher serum β-HCG concentrations than the MTX group. This indicates that at least some of them were not good candidates for MTX treatment.

It is concluded that prior MTX treatment or salpingectomy does not affect ovarian response in the subsequent IVF cycle.

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