Failure to improve a thin endometrium in the late proliferative phase with uterine infusion of granulocyte-colony stimulating factor

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Summary

Purpose: To determine if the treatment with uterine infusion of granulocyte colony-stimulating factor (G-CSF) can improve endometrial thickness in an infertile woman with a double uterus, who consistently showed a thin endometrium in the late proliferative phase either in controlled ovarian hyperstimulation (COH) IVF-ET cycles or with graduated estrogen/sildenafil protocols for frozen embryo transfer (ET). *Materials and Methods:* A single uterine infusion of G-CSF was performed in the late proliferative phase in a woman who only attained a five-mm thickness despite a high dose vaginal and oral estradiol regimen plus sildenafil. *Results:* No increase was found within a couple days. *Conclusions:* A previous four-case study in another center found 100% improvement in the endometrial thickness in women with consistently thin endometria. Perhaps the uterine anomaly in the present case prevented the response of the endometrium.

Key words: Granulocyte colony-stimulating factor; Thin endometrium; In vitro fertilization-embryo transfer; Intrauterine infusion.

Introduction

Lower pregnancy rates (PRs) per embryo transfer (ET) have been demonstrated in women with thin endometria in the late proliferative phase at the time of human chorionic gonadotropin (hCG) injection [1,2]. One review of the literature concluded that there were no successful pregnancies following in vitro fertilization-embryo transfer (IVF-ET) when the pre-ovulatory endometrium was < six mm [3]. However subsequent to this study, a successful pregnancy following IVF was reported where the maximum endometrial thickness was only four mm [4]. A successful delivery was also reported without IVF-ET in a natural cycle with a maximum endometrial thickness in the late proliferative phase of four mm [5].

A study of 35 women having IVF-ET with a maximal thickness of ≤ five mm found three clinical pregnancies (8.5% per transfer) and two live deliveries (5.7% per transfer) [6]. In that same study there was one live delivered pregnancy out of seven frozen ETs (14.2%) with maximal endometrial thickness ≤ five mm.

Most therapies to improve thin endometria have failed. Though at one time treatment during the follicular phase with vaginal sildenafil seemed promising, other studies failed to confirm any significant beneficial effect on pregnancy rates [7,8].

Another promising treatment for unresponsive thin endometrium has recently been published, i.e., the uterine perfusion of granulocyte colony-stimulating factor (G-CSF) [9]. They reported that four consecutive women with a history of multiple cycles of failing to attain an adequate endometrial thickness despite the use of higher dose graduated estradiol and vasodilators resulting in cancellation of ET cycles [9]. However, with intrauterine infusion of G-CSF, three improved the endometrial thickness to eight to ten mm and one increased to 7.3 mm [9]. Even more important, all four conceived and three delivered live babies [9].

The authors published these cases ahead of two ongoing prospectively randomized studies of G-CSF "because of unexpectedly clear results in clinical circumstances without effective treatment options". They suggested that this small study could influence other centers to try this therapy and hopefully improve their success rates prior to the publications of the results of the large prospective study.

The present case report is the authors' first attempt to improve the endometrial thickness of a woman with multiple unresponsive cycles using G-CSF. Unfortunately they present their first case of failure with this technique.

Case Report

This 33-year-old woman had multiple ETs but failed to conceive. The failures were attributed to inadequate endometrial thickness on the day of hCG. She had the remaining frozen embryos transferred to the present IVF center. However despite vaginal and oral estradiol extending to day 10 and vaginal sildenafil 25

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mg 4x/day from day 7, her maximal endometrial thickness was six mm. She failed to conceive. She then had controlled ovarian hyperstimulation (COH) for IVF-ET starting with 225 IU highly purified follicle stimulating hormone (FSH) and 75 IUI highly purified menotropins. Despite attaining 23 follicles of adequate size for oocyte retrieval, her endometrial thickness was only a six mm triple line pattern on the day of hCG. She failed to conceive following two ETs. A milder stimulation protocol was used in her next frozen ET cycle and she reached a maximum seven-mm endometrial thickness. She then did another COH IVF-ET cycle and reached a maximum thickness of six mm.

Based on the aforementioned study by Gleicher *et al.*, the present authors elected to add to the estrogen sildenafil protocol uterine perfusion G-CSF which was performed according to the described technique by Gleicher *et al.* [9]. Unfortunately this therapy failed to increase the five-mm endometrial thickness at all. A blastocyst transfer was performed on day 5 and the pregnancy test was negative.

Discussion

Other than the aforementioned cases described of an IVF-ET live pregnancy achieved with a four-mm endometrial thickness, here have been other anecdotal reports of successful pregnancies with very thin endometria [4, 5]. One case was recently reported of a woman with marked diminished oocyte reserve who conceived with her own oocyte following a fresh embryo transfer using a mild ovarian hyperstimulation regimen [10]. Interestingly she had been enrolled in a European IVF center for donor oocytes but after two attempts to improve endometrial thickness with estrogen and then estrogen plus vasodilators, she was advised that they would only transfer the embryos to a gestational carrier. Obviously if she would have used G-CSF (which she did not) the conclusion would have been that the G-CSF must have been responsible for the successful implantation since there had never been one precedent for a pregnancy with IVF or natural with an endometrial thickness < four mm.

There has also been a report of a successful twin pregnancy in a donor oocyte recipient whose maximum endometrial thickness was four mm [11].

The particular woman in the present case report had a double uterus and a vaginal septum. Possibly the uterine anomaly was responsible for the failure to respond to the G-CSF in this case and hopefully G-CSF will prove to be an effective therapy for improving pregnancy rates following ET with thin endometria or prove to be an effective agent to improve implantation rates even if there seems to be adequate endometrial thickness.

The report from Gleicher *et al.* is not the first to suggest that G-CSF can improve the implantation potential and reduce miscarriage rates [12, 13]. Granulocyte-CSF has even been added to endometrial co-culture cells as early as 1998 [14]. What is unique about the procedure proposed by Gleicher *et al.* is the use of G-CSF by uterine infusion.

There is no question that although a very thin endometrium is not an absolute factor that prevents successful implantation, there is no question that very thin endometria negatively effects pregnancy outcome following ET. By adding the present case report to the other four, the use of G-CSF is 80% effective. Hopefully the large prospective study that is underway will show an 80% efficacy rate. One just has to be a little careful about conclusions from a study being performed with a patented procedure by a group who benefits from approval. Thus the present authors agree with Gleicher *et al.* that other centers should independently conduct controlled studies. The problem is that the percentage of women who cannot stimulate a thickness past five mm is low and it may be that these two case reports should encourage more anecdotal reports pro or con to get an early determination of the efficacy of G-CSF for increasing the endometrial thickness.

Of course the use of G-CSF need not be restricted to cycles involving ET. For the aforementioned patient, the authors considered intrauterine infusion of G-CSF even in natural cycles to see if it could improve implantation even if it did not improve endometrial thickness. She elected not to take the G-CSF. Two months later she conceived in a natural cycle. Her only treatment is progesterone which she will continue through her first trimester. Had she decided to continue on monthly G-CSF therapy, who would question that the successful pregnancy was not related to the G-CSF treatment. Instead it is clear that the pregnancy had nothing to do with G-CSF therapy.

Gleicher *et al.*, in their manuscript mentioned that they are in the midst of a prospective randomized study to evaluate G-CSF on thickness and implantation. The authors' plans are to evaluate G-CSF in a small series in an attempt to corroborate or refute the effect of G-CSF for thin endometria.

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