

# Does Acupuncture Improve the Endometrium for Women Undergoing an Embryo Transfer: A Pilot Randomised Controlled Trial

**Dr Caroline Smith**\*<sup>1,2</sup> PhD

**Dr Meaghan Coyle**<sup>3</sup> PhD

**Prof Robert J Norman**<sup>1,4</sup> MD

1. Centre for Reproductive Health, School of Paediatrics and Reproductive Health, The University of Adelaide, Australia

2. CompleMED, The University of Western Sydney, Australia

3. Private practice, Sale, Victoria, Australia

4. Repromed, Adelaide, Australia

## ABSTRACT

**Background:** There is a growing body of research suggesting acupuncture may increase pregnancy and live births, when administered on the day of embryo transfer. The physiological effects of acupuncture that may influence the outcome from embryo transfer remain unclear. **Aims:** To examine the effects of acupuncture on uterine endometrium thickness and pattern, and the level of hormonal medication during an IVF cycle. **Design:** Randomised controlled trial. **Subjects:** Women undergoing an IVF cycle with a planned embryo transfer at day 3 or day 5. **Setting:** A reproductive medicine unit in South Australia. **Intervention:** Women were randomly allocated to acupuncture or standard care. Women in the acupuncture group received three treatments, the first undertaken on day 9 of stimulating injections, and two on the day of embryo transfer. **Outcome measures:** The primary outcomes were change in endometrial thickness and pattern, and levels of plasma progesterone, and oestradiol during the IVF cycle, through to seven days post-egg retrieval. Secondary outcomes included number of oocytes retrieved, number of oocytes fertilised, and biochemical pregnancy rate. **Results:** Endometrial thickness and pattern and levels of hormonal medication did not differ between groups on the day of embryo transfer or in the luteal phase ( $p > 0.05$ ). There were no differences in any secondary outcomes. **Discussion and Conclusion:** The results of this pilot study suggest acupuncture did not influence the endometrium or levels of hormonal medication during the IVF cycle. The small number of subjects and incomplete data make conclusions difficult, and consideration must be given to whether the measurement parameters were sensitive to changes from acupuncture, or whether the study sample was too small to detect a change. Interestingly, for the women who received acupuncture, there was a non-significant trend towards a higher fertilisation rate and numbers of women proceeding to embryo transfer.

**KEYWORDS** acupuncture, infertility, randomised controlled trial.

\* Correspondent author; e-mail: caroline.smith@uws.edu.au

## Introduction

There is a small but growing body of research examining the effectiveness of acupuncture as an adjunct to in-vitro fertilization (IVF). There are two systematic reviews reporting on a benefit from acupuncture when it is used as an adjunct on the day of embryo transfer (ET) for women undergoing assisted reproductive technology (ART) (OR 1.65, 95% CI 1.27 to 2.14).<sup>1</sup> A recent Cochrane review reported evidence of benefit when acupuncture is performed on the day of ET on the live birth rate (OR 1.89, 95% CI 1.29 to 2.77).<sup>2</sup> However, further research is required.

The endometrium in IVF cycles is subject to an altered endocrine environment, and factors influencing endometrium receptivity are poorly understood.<sup>3</sup> The thickness of the endometrium, morphology and uterine artery blood flow however have been identified as potential, important variables influencing the success of the implantation by the embryo.<sup>4</sup> Endometrium thickness is required to optimize the pregnancy rate and is partly a reflection of uterine artery blood flow. Pregnancies generally occur where the endometrium achieves a thickness of greater than 8 mm.

Conventional science has hypothesised that acupuncture works by neurological, neuro-hormonal and psychological mechanisms. Needle insertion into the skin and deeper tissues results in a particular pattern of afferent activity in peripheral nerves. The mechanisms through which acupuncture might influence female fertility could involve central effects on the release of neurotransmitters including  $\beta$ -endorphin, which in turn influence gonadotropin releasing hormone (GnRH) release and impact on pituitary gonadotropin secretion, ovarian follicular growth, ovulation and fertility.<sup>5-7</sup> Acupuncture may also exert a sympatho-inhibitory effect which may reduce uterine artery impedance and thereby increase uterine and ovarian blood flow. Stener-Victorin (1996) demonstrated that when acupuncture was administered to 10 infertile women, blood flow impedance was reduced in women receiving acupuncture ( $P < 0.05$ ).<sup>8</sup>

It has been suggested that this improved uterine blood flow could improve the growth and thickness of the endometrium. Acupuncture might also impact on local humoral factors that are involved with the regulation of implantation. A study by Paulus however found no difference between groups on the endometrium thickness and plasma oestradiol on the day of transfer.<sup>9</sup> The pulsatility index of uterine arteries before and after transfer was also not found to differ between groups ( $p > 0.05$ ). In another trial, Quintero evaluated the effectiveness of acupuncture as an adjunct to IVF and found acupuncture significantly reduced the amount of gonadotropin used ( $p < 0.05$ ), however no effect on the pregnancy rate was found.<sup>10</sup>

The physiological effects of acupuncture that may influence the outcome from embryo transfer remain unclear. There is currently no data available on whether acupuncture treatment administered earlier in the cycle effects the growth of the endometrium over the subsequent days following embryo transfer. The aim of this research was to examine a potential mechanism arising from acupuncture that may affect endometrial receptivity and subsequently increase the success of pregnancy rates. The research examined the effects of acupuncture on endometrium thickness and pattern from baseline, at embryo transfer and in the luteal phase, and the level of hormonal medication (plasma progesterone, and plasma oestradiol) during the IVF cycle through to seven days post-egg retrieval. The study also aimed to pilot the processes and procedures for undertaking further research in this area.

## Methods

### PARTICIPANTS AND METHODS

We recruited women to this trial from Repromed, a reproductive medicine unit in Adelaide, South Australia between October and December 2007. The research was approved by the Women's and Children's Hospital research and ethics committee, Adelaide. Women undergoing an IVF cycle with a planned embryo transfer at day 3 or day 5 were eligible to join the study. Women who were planning a frozen embryo transfer, or who were planning to have acupuncture were excluded from the study. Eligible women were identified by a research nurse at Repromed. All women received hormonal stimulation as per Repromed treatment protocols.

### INTERVENTION

Women were recruited to the trial on day 9 of their IVF cycle. After obtaining their written consent, subjects underwent a traditional Chinese medicine (TCM) differential diagnosis to identify the predominant TCM syndrome, and baseline data was collected. Women were randomised to acupuncture or standard care groups. Randomisation was undertaken by the study acupuncturist, by taking the next sequentially numbered sealed opaque envelope. An independent researcher developed the computer generated randomisation schedule. The number of cases in each randomisation block was 2 and 4 and this was not revealed to the acupuncturist.

The two acupuncturists who administered the acupuncture over the study period were accredited members of the Australian Acupuncture and Chinese Medicine Association Ltd. Both acupuncturists have previous experience in this area, and with administering acupuncture clinical trials and private practice, (CS four years training and fifteen years clinical experience, MC four years training and nine years clinical experience). The majority of women saw the same acupuncturist for all

treatments. The trial was administered in private acupuncture clinic rooms, a 10 minute drive from the fertility centre. For women receiving acupuncture, an individualised acupuncture treatment was administered on day nine of the IVF cycle, as per our previous trial.<sup>11</sup> Treatment was administered according to TCM pattern differential diagnosis, and in consideration of the 'stimulation' stage of the IVF cycle, for example, supporting Kidney yin and building blood. The second and third acupuncture treatments were administered on the day of embryo transfer, and administered before and after the embryo transfer. The acupuncture treatment administered prior to embryo transfer included points PC6 *Neiguan*, SP8 *Diji*, LR3 *Taichong*, ST29 *Guilai*, and CV4 *Guanyuan*, and auricular acupuncture points *Shenmen*, *Zhigong*, *Neifenmi* and *Naodian*. The treatment administered after embryo transfer included acupuncture points ST36 *Zusanli*, SP6 *Sanyinjiao*, and SP10 *Xuehai*, and auricular acupuncture points *Shenmen*, *Zhigong*, *Neifenmi* and *Naodian*. Modifications were made to the Paulus treatment protocol<sup>9</sup>; we substituted CV4 *Guanyuan* for CV6 *Qihai*, to provide greater support to the Kidney qi. We excluded LI4 *Hegu*, due to its use contra-indicated in early pregnancy, and to promote menstruation<sup>12</sup> and GV20 *Baihui* was excluded due to the multiple use of acupuncture points to calm the patient.<sup>9</sup> Acupuncture was applied bilaterally, with the exception of the four auricular acupuncture points used on the day of transfer. Before the transfer, two points were needled in the right ear, and the other two points were needled in the left ear. After embryo transfer the side of auricular acupuncture was reversed. Seirin brand 0.22 x 30 mm acupuncture needles were inserted to tissue level and stimulated manually to elicit the *deqi* response. Needles were retained for 25 minutes in each treatment.

Women randomised to the standard care group received the standard Repromed treatment protocols only. Women allocated to this group were also offered acupuncture at no cost if they were to undergo a future embryo transfer.

Primary outcome data was assessed from ultrasound and blood tests. An assessment of the endometrium was made recording the pattern and thickness of endometrium by vaginal ultrasound. The ultrasound was performed by a nurse at the clinic who was blind to the subject's study group. A routine ultrasound scan was undertaken at day nine to assess timing for egg retrieval. Further ultrasound measurements were undertaken at the time of embryo transfer, and an additional non-routine ultrasound was undertaken one week following egg retrieval. The endometrial pattern was categorised by the study nurse blind to group allocation. Classification of endometrium pattern was made based on three criteria proposed by Gonen and Casper<sup>12</sup> as follows: type A, an entirely homogenous, hyperechogenic pattern without a central echogenic line; type B, an intermediate isoechogenic pattern, with the same

reflectivity as the surrounding myometrium and a poorly defined central echogenic line; or type C, a multilayered 'triple-line' endometrium, consisting of a prominent outer and central hyperechogenic line and inner hypoechogenic region. Routine blood collection was undertaken on day nine of the IVF cycle for plasma oestradiol and progesterone, and additional blood was taken on day seven following egg retrieval. Nurses taking the blood were blind to the woman's group allocation. Secondary outcomes included number of oocytes retrieved, number of oocytes fertilized, and biochemical pregnancy rate, and were collected from clinical case records.

This was a pilot study, there was no data reported on the thickness or pattern of the endometrium in the literature to guide a sample size power calculation. The aim of the study was therefore to have 20 subjects available for analysis. The initial analysis examined the baseline characteristics of women randomised to the trial. The main analyses used an 'intention to treat' approach and compared differences in the primary study outcome measures between the two groups over time using repeated-measures analysis of variance. Comparisons were also made between groups in binary variables using the chi-square test. Data on serum was analysed using the Mann Whitney test. The analysis was undertaken by the investigator blind to study group. Levels of significance were reported at  $p < 0.05$ . Data were analysed using SPSS version 11.5.

## Results

Forty six women were approached to participate in the trial (Figure 1). Eighteen women declined to participate in the trial, the reasons given included not wishing to be randomised to the study, the woman was already having acupuncture, or they were unable to attend for all proposed acupuncture sessions. Twenty eight women agreed to be randomised to the trial, and fourteen women were allocated to each group. One woman was withdrawn from the trial when she was unable to attend for measurements of the primary endpoints following randomisation. Six women were unable to complete their participation in the trial due to their treatment not progressing to embryo transfer; five of these women were in the control group, and one woman was in the acupuncture group.

The mean age of women participating in the trial was 35 years (Table 1). The majority of women had completed at least one IVF cycle, had a body mass index just above the normal range, and 64% were childless. Over 64% of women had experienced infertility for greater than two years, and the main reasons for infertility were unexplained (25%), or male factor (28%). Almost all women had finished high school (96%), 92% had completed vocational training award or university degree, and 86% were employed outside the home. Fifty percent had used

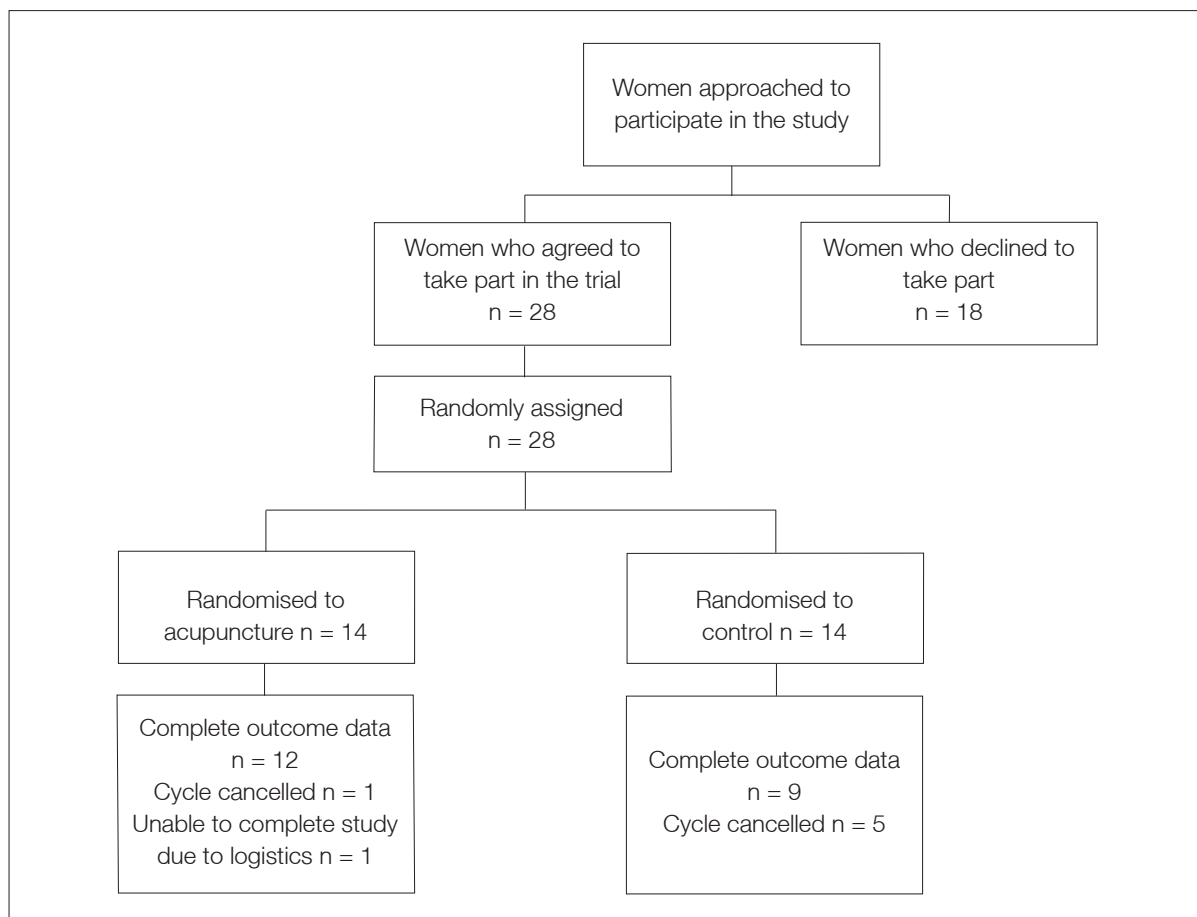


FIGURE 1 Trial flow chart

acupuncture previously. No differences existed between groups, suggesting randomisation produced comparable groups at baseline. We also present details of women’s TCM diagnosis by group (Table 2). A Kidney deficiency was diagnosed in over 50% of women. The most frequent diagnosis was Kidney yang, and Qi stagnation. There were no differences in TCM diagnosis between groups ( $p = ns$ ).

The primary and secondary outcomes are presented in Table 3. Endometrial thickness did not differ between groups at baseline, the day of embryo transfer or in the luteal phase ( $p > 0.05$ ). Data on outcomes at three time-points were available from nine women; however, there was no difference in endometrial thickness or pattern between groups over time. There were no differences found between groups for other study outcomes: plasma progesterone and oestradiol levels at baseline and 7 days post-egg retrieval, number of oocytes retrieved, number of oocytes fertilized, and biochemical pregnancy rate.

## Discussion

The findings from this pilot study provide no preliminary results that acupuncture administered prior to and on the day of embryo transfer has an influence on endometrial thickness or pattern, or plasma levels of progesterone and oestradiol levels. However, the small numbers of subjects, and incomplete data make conclusions difficult, and consideration must be given to whether the current measurement parameters were sensitive to changes from acupuncture, and that the pilot sample size was too small to demonstrate any small changes in the primary endpoints. We can conclude that the effect of acupuncture on these outcomes is not large. We were unable to measure blood flow in this study and the degree of endometrium vascularisation remains unknown. Interestingly, for the women who received acupuncture, there was a non-significant trend towards a higher fertilisation rate and the number of women proceeding to embryo transfer. Although not an outcome of the study the pregnancy rate was also not found to differ between groups, and may be explained by the small sample.

TABLE 1 Characteristics of the women at trial entry

	Acupuncture n = 14	Control n = 14
Age (years)	36.0 (5.1)	35.7 (5.0)
No. of previous IVF cycles		
0	2 (14.3)	3 (21.4)
1–3	10 (71.4)	6 (42.9)
4+	2 (14.3)	5 (35.7)
Duration of infertility (years)		
<2 y	5 (35.7)	5 (35.7)
2–4	9 (64.3)	7 (50.0)
5+	0 (0.0)	2 (14.3)
Parity		
0	7 (53.8)	11 (78.6)
1+	6 (46.2)	2 (21.4)
Reason for infertility		
Male factor	4 (28.6)	4 (28.6)
Maternal age	4 (28.6)	3 (21.4)
Tubal	1 (7.1)	1 (7.1)
Unexplained	4 (28.6)	3 (21.4)
Endometriosis	0 (0)	1 (7.1)
BMI	26.1 (4.3)	23.9 (4.8)
Finished high school	13 (100)	13 (92.9)
Completed tertiary education	12 (92.3)	13 (92.9)
Employed outside the home	14 (100)	13 (92.9)
Previous use of acupuncture	7 (50.0)	7 (50.0)

Values are number (%) of women or mean (SD)

TABLE 2 TCM diagnosis by group

	Acupuncture n = 14	Control n = 14
Kidney yang	8 (57.1)	6 (42.9)
Kidney yin	5 (35.7)	5 (35.7)
Blood deficiency	1 (7.1)	3 (21.4)
Heat	2 (14.3)	0 (0.0)
Qi stagnation	8 (57.6)	6 (42.9)
Blood stagnation	3 (21.4)	3 (21.4)
Sp Qi deficiency	5 (35.7)	3 (21.4)

Values are number (%) of women

Resources for the trial were to allow data analysis from 20 women. Although resources were allocated to 28 women randomised to the study, seven women withdrew due to their IVF cycle being cancelled. We also encountered some logistical difficulties with obtaining measurements of the endometrium. For embryo transfers occurring at the weekend we did not have access to staff who would undertake measurements for the study. We also found for those women proceeding to embryo transfer, some were reluctant to return for their blood and ultrasound measurements. These are important factors identified from the pilot relating to feasibility and study design, which will guide researchers with planning future research in this area.

One potential limitation of the study was the times scheduled for measurement of the endometrium and hormonal measurements. We scheduled measurements following embryo transfer (day 3 or day 5), and day seven, post-egg retrieval. This was based on an assumption that any acupuncture effect would be detectable at these time points. Although the day of embryo transfer varied between day 3 or day 5, both groups of women would have received hormonal stimulation to ensure the endometrium thickness met the IVF centre criteria. The current literature provides no evidence for when, and for how long, biophysical changes may occur after acupuncture; it is possible that any changes that may have occurred between and during the test interval have escaped detection. A study design with frequent measurements after initial exposure and longer intervals between measurements as time passes, may allow for detection of any acupuncture effects; however, the intensive requirements of this type of design may not be acceptable and practical for women receiving IVF treatment.

While there is a growing body of evidence for acupuncture improving the pregnancy rate with IVF treatment, the mechanism of action remains elusive. Research examining the potential mechanism of acupuncture following embryo transfer has focused on measuring uterine blood flow and changes within the endometrium. These preliminary studies suggest no effect from acupuncture. The aims of acupuncture treatment when administered on the day of embryo transfer are to increase blood and energy flow to the uterus, to sedate the patient and to stabilize the endocrine system.<sup>9</sup> Other potential mechanisms should be given consideration. Several papers have suggested that psychological state can impact on the chance of success with an IVF cycle.<sup>12</sup> There is little research examining the effect of acupuncture on the psychological state (either through

TABLE 3 Primary and secondary outcomes by study group

	N	Acupuncture	Control	Significance p value
Endometrial thickness				
Baseline	12/9	9.9 (1.8)	9.7 (1.4)	0.89
Embryo transfer	9/3	11.5 (2.0)	11.1 (2.0)	0.79
7 days post-egg retrieval	11/5	11.9 (3.0)	14.6 (3.7)	0.14
Endometrium pattern				
Baseline	10/8			0.63
B		4 (40.0)	5 (62.5)	
C		2 (20.0)	1 (12.5)	
B-C		4 (40.0)	2 (25.0)	
7 days post-egg retrieval	11/5			0.48
B		10 (90.9)	5 (100.0)	
C		1 (9.1)		
Oestradiol nmol/L				
Baseline	12/10	2.5 (1.9) (11.71)	2.2 (1.1) (11.25)	0.87
7 days post-egg retrieval	12/5	2.4 (1.7) (9.29)	2.2 (1.4) (8.30)	0.72
Progesterone nmol/L				
Baseline	11/11	3.2 (0.7) (12.36)	3.5 (2.2) (10.64)	0.56
7 days post-egg retrieval	12/5	389.6 (6.2) (9.17)	313.4 (221.4) (8.6)	0.87
Number of oocytes retrieved per subject	13/11	8.9 (6.2)	8.4 (5.6)	0.85
Number of oocytes fertilised per subject	12/11	4.5 (4.4)	2.4 (2.4)	0.19
Biochemical pregnancy	12/9	2 (16.7)	2 (22.2)	0.74

Values are number (%) of women or mean (SD) with mean rank

biophysical measures or self-report questionnaires) of women undergoing IVF. Research in these areas, for example, cortisol levels as an indicator of relative levels of stress, may provide more information about possible mechanisms of acupuncture.

There is a need for further research in this area to clarify possible mechanisms of acupuncture. It is recommended that clinical trials incorporate collection of other biophysical and psychological measures to further the knowledge base.

### Conflict of interest statement

The authors have no conflict of interest.

### Acknowledgements

We wish to thank the women participating in the trial, our thanks also to the Repromed staff for their assistance with recruitment and implementation, and in particular Paula Scanlon for undertaking the ultrasounds, and Helen Alvino, research manager, for her assistance with trial implementation.

Funding for this study was provided from a grant made by the Australian Acupuncture and Chinese Medicine Association Ltd.

Clinical trial registration

<[www.anzctr.org.au](http://www.anzctr.org.au)> registration number 12607000015448.

## Clinical Commentary

IVF is a common and accepted form of treatment for many couples seeking assistance with achieving a pregnancy. Acupuncture may increase clinical pregnancy and live birth rates when administered on the day of embryo transfer, and interest is turning to exploring potential mechanisms. Our small pilot feasibility study found insufficient evidence to suggest acupuncture had an effect on the endometrium and serum levels of oestradiol and progesterone. The clinical implications of this study provide practitioners with no insight to a possible mechanism. The study does assist clinical researchers working in this discipline with the design and planning of future research.

## References

1. Manheimer E, Zhang G, Udoff L, Haramati A, Langenberg P, Berman BM, et al. Effects of acupuncture on rates of pregnancy and live birth among women undergoing in vitro fertilization: a systematic review and meta-analysis. *BMJ* 2008;336(7643):545–9.
2. Cheong YC, Hung Yu Ng E, Ledger WL. Acupuncture and assisted conception. *Cochrane Database Syst Rev.* 2008 8;(4):CD006920.
3. Borgain C, Devroey P. The endometrium in stimulated cycles for IVF. *Hum Reprod Update* 2003;9(6):515–22.
4. Schild RL, Knobloch C, Dorn C, Fimmers R, van der Ven H, Hansmann M. Endometrial receptivity in an in vitro fertilization program as assessed by spiral artery blood flow, endometrial thickness, endometrial volume, and uterine artery blood flow. *Fertil Steril* 2001;75(2):361–6.
5. Ferin M, Vande Wiele R. Endogenous opioid peptides and the control of the menstrual cycle. *Eur J Obstet Gynecol Reprod Biol* 1984;18(5–6):365–73.
6. Petraglia F, Di Meo G, Storchi R, Segre A, Facchinetti F, Szalay S, et al. Proopiomelanocortin-related peptides and methionine enkephalin in human follicular fluid: changes during the menstrual cycle. *Am J Obstet Gynecol* 1987;157(1):142–6.
7. Chang R, Chung PH, Rosenwaks Z. Role of acupuncture in the treatment of female infertility. *Fertil Steril* 2002;78(6):1149–53.
8. Stener-Victorin E, Waldenström U, Andersson SA, Wikland M. Reduction of blood flow impedance in the uterine arteries of infertile women with electro-acupuncture. *Hum Reprod* 1996;11(6):1314–17.
9. Paulus WE, Zhang M, Strehler E, El-Danasouri I, Sterzik K. Influence of acupuncture on the pregnancy rate in patients who undergo assisted reproduction therapy. *Fertil Steril* 2002;77(4):721–4.
10. Quintero R. A randomized, controlled, double-blind cross-over study evaluating acupuncture as an adjunct to IVF. *Fertil Steril* 2004;81(S3):S11–12.
11. Smith C, Coyle M, Norman RJ. Influence of acupuncture stimulation on pregnancy rates for women undergoing embryo transfer. *Fertil Steril* 2006;85(5):1352–8.
12. Betts D. Essential guide to acupuncture in pregnancy and childbirth. Hove, UK: The Journal of Chinese Medicine; 2006.
13. Gonen Y, Casper RF. Prediction of implantation by the sonographic appearance of the endometrium during controlled ovarian stimulation for in vitro fertilization (IVF). *J In Vitro Fert Embryo Transf* 1990;7(3):146–52.
14. Eugster A, Vingerhoets AJJM. Psychological aspects of in vitro fertilization: a review. *Soc Sci Med* 1999;48(5):575–89.