子宮內膜血流對胚胎著床之影響

Intra-endometrial vascularity in embryo implantation

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主持人：楊友仕
執行機構及單位名稱：台大醫學院婦產科

一、中文摘要

多數學者均認為體外受精週期中，子宮內膜厚度大或等於 10 mm **有利於胚胎著床，但是他們無法解釋為何有些婦女具有足夠的內膜厚度，卻始終無法達到成功的胚胎著床。本研究計畫中，我們嘗試利用能量超音波定量子宮內膜之血流，稱為子宮內膜血流面積** (EPDA)**，以之作為厚度之外的另一內膜接受度判斷標準，並比較其與懷孕預後的關聯性。我們挑選體外受精週期中子宮內膜厚度** ≥10 mm **的婦女，在** 95 **個研究對象中, 懷孕率與胚胎著床率均隨著內膜血流面積的增加而上升，而且在** EPDA = 5 mm^2 **時達到高峰, 因此我們以** EPDA = 5 mm^2 **為臨界點將** 95 **位婦女分為二組。較諸對照組，內膜血流面積較大的婦女** (EPDA ≥5 mm^2) **可以達到較高的懷孕率** (47.5% vs. 23.5%, p = 0.021) **與胚胎著床率** (20.2% vs. 8.1%, p = 0.003)**。本研究結論為：子宮內膜血流面積是除了內膜厚度之外，另一影響內膜接受度的重要因素。當婦女內膜厚度足夠，但血流貧乏時，對其懷孕預後將有不良影響。此結果在臨床上的可能應用包括：**(1) **依照內膜血流灌注情形決定胚胎植入的數目。對於內膜血流貧乏的婦女植入較多的胚胎以提高懷孕機率；而針對內膜血流豐富的婦女植入較少的胚胎以降低多胞胎機率。**(2) **驗證一些藥物如** aspirin**、** heparin **及** nitroglycerine **其促進胚胎著床率的機轉是否由於改善了內膜血流的灌注。**

二、英文摘要

An endometrial thickness of 10 mm or more has been reported to be favorable for embryo implantation in in-vitro fertilization (IVF) programs. Nevertheless, many women, although having adequate endometrial thickness, have been unable to achieve satisfactory implantation. With the aid of power Doppler sonography, we attempt to test the association of intra-endometrial vascularity and reproductive outcome. In this study, we only enrolled women who had an endometrial thickness ≥ 10 mm. This group was made up of 95 women undergoing IVF cycles, which resulted in 37 intrauterine pregnancies. Those who had apparent endometrial pathologies were excluded from the study. Among women with similar ages, body mass indices, peak estradiol levels, endometrial thickness, and after transferring a similar number of embryos, those women having cycles with an intra-endometrial power Doppler area (EPDA) < 5 mm^2 achieved a significantly lower pregnancy rate (23.5% vs. 47.5%, p = 0.021) and implantation rate (8.1% vs. 20.2%, p = 0.003) than those with an EPDA ≥ 5 mm^2. We conclude that, in addition to endometrial thickness, EPDA may serve as a valuable factor that was closely associated with endometrial receptivity. Women having adequate endometrial thickness but a small EPDA were correlated with an unfavorable reproductive outcome.

三、計畫緣由與目的

Embryo quality and endometrial

receptivity are two final parameters which determine the reproductive outcome in in-vitro fertilization (IVF) programs. Morphological assessment of the embryos using light microscopy is a generally accepted method for the evaluation of embryo quality. However, the evaluation of endometrial receptivity remains challenging in clinical practice. In the attempt to identify a non-invasive uterine predictor of endometrial receptivity during IVF procedures, sonographic imaging of endometrial patterns and thickness has been widely studied. Some investigators have found (Dickey et al., 1992, Noyes et al., 1995) and others have not found (Khalifa et al., 1992, Oliveira et al., 1993) a positive correlation between endometrial echo patterns and pregnancy rates. On the other hand, cycles with an endometrial thickness of 10 mm or more have been reported to be favorable for embryo implantation (Check et al., 1991, Rinaldi et al., 1996), although other cutoff values have also been claimed as appropriate (Dickey et al., 1992, Noyes et al., 1995). However, these reports failed to explain why some women had adequate endometrial thickness, but could not achieve a satisfactory implantation rate.

Goswamy and Steptoe (1988) first reported that impaired perfusion of the uterine arteries may be a cause of infertility and that it can be related to unsuccessful in-vitro fertilization treatment. Utilizing transvaginal color Doppler ultrasound, some authors have been able to distinguish between conception and non-conception cycles in IVF patients before the embryo transfer. These researches were based on the differences in mean uterine artery pulsatility index (PI) or resistance index (RI). Patients who had become pregnant had a lower vascular impedance than those who had not (Sterzik et al., 1989, Steer et al., 1992 and 1995). However, recent data have challenged the earlier assumptions about the predictive role of Doppler measurements in assisted reproductive technology (ART) procedures (Bassil et al., 1995, Tekay et al., 1996, Schwartz et al., 1997). This may be due to the nature of uterine artery Doppler measurements, which are based on flow to the entire uterus, not to focal areas of the endometrium. For successful implantation of an embryo, the quality of the endometrium may be more important than the global blood flow throughout the uterus (Schwartz et al., 1997). Zaidi et al. (1995) utilized color Doppler imaging and found that absent subendometrial and intraendometrial vascularization was associated with implantation failures in IVF cycles. However, the sensitivity of color Doppler is limited and thus the rather weak vascular signals inside the endometrium were difficult to be captured.

Power Doppler sonography, with the advantages of less direction dependence, higher sensitivity, and better contrast of vascular contour, is a favored tool in studies of tissue and tumor vascularity (Taylor et al., 1996). Compared to color Doppler, power Doppler has been claimed to be more sensitive to low flow states (Jain et al., 1991). In addition to qualitative studies, computer-assisted quantitative assessment of power Doppler vascular signals has been made possible and applied preliminarily in studies of breast tumors (Huber et al., 1994, Kedar et al., 1995) and cervical lymphadenopathies (Wu et al., 1998). It was explained as discriminating clearly between benign and malignant tissues because of their different densities in vascularity. Utilizing the power Doppler sonography, we were able to obtain an in vivo quantification of macroscopic vascularity in the endometrium, an area containing rather weak vascular signals that could hardly be captured with conventional color Doppler.

The aim of this study was to search for another factor that, in addition to endometrial thickness, might affect endometrial receptivity. With the aid of power Doppler sonography, we set up a method to measure the intra-endometrial power Doppler area (EPDA), and to examine its association with the following IVF outcome.

四、結果與討論

In this study, 95 women underwent their IVF cycles, resulting in 43 pregnancies. The etiologies of infertility were tubal factor in 28 cycles, ovarian factor in 6, male factor in 21, endometriosis in 6, combined factor in 23, and unexplained infertility in 11. After subtracting 3 biochemical and 3 tubal pregnancies, 37 intrauterine pregnancies were achieved. The EPDA of the 95 IVF cycles ranged from 0.4 to 39.5 mm^2, with a median area of 6.3 mm^2 (interquartile range 4.2 to 11.5). Women who achieved intrauterine pregnancies had a significantly larger EPDA than those who did not (8.8 mm^2 (5.1 to 16) vs. 5.8 mm^2 (4 to 9.3), $p < 0.02$). The lowest EPDA in a cycle resulting in pregnancy was 1.1 mm^2

The EPDA in relation to pregnancy and implantation rates was analyzed. The pregnancy and implantation rates were higher for women with a larger EPDA. The implantation rates in women having cycles with an EPDA between 5 and 8, between 8 and 11, and larger than 11 mm^2 were all significantly higher than those with an EPDA < 5 mm^2 (25.9%, 17.6% and 17.4% respectively, vs. 8.1%, $p < 0.05$). Therefore, we assumed an EPDA of 5 mm^2 as the threshold, and an area < 5 mm^2 was regarded as poor vascularization.

No significant differences in age and body mass index (BMI) existed between women having an EPDA ≥ 5 mm^2 and those having an EPDA < 5 mm^2. Peak estradiol levels and the numbers of oocytes were not different between the two groups. After the transfer of a similar number of embryos, women having cycles with an EPDA ≥ 5 mm^2 achieved a significantly higher pregnancy rate (47.5% vs. 23.5%, $p = 0.021$) and implantation rate (20.2% vs. 8.1%, $p = 0.003$) than those having cycles with an EPDA < 5 mm^2. The multiple pregnancy rates were 48.3% and 12.5% respectively, for women with a larger and smaller EPDA. The rates of spontaneous abortion were similar between women containing larger and smaller EPDA (24% vs. 38%). If we took fetal viability (> 20 weeks) into account, the association of EPDA and implantation rates was strengthened (17.8% vs. 5.6%, $p = 0.001$).

Even with similar endometrial thicknesses (12.9 ± 2.6 vs. 12.0 ± 1.7 mm) as well as mean uterine artery PI (1.65 ± 0.22 vs. 1.67 ± 0.21) and PSV (35.9 ± 8.9 vs. 35.0 ± 10.1 cm/sec), there were different manifestations of intra-endometrial vascularity.

Endometrial thickness of 10 mm or more has been generally regarded as a favorable factor for implantation. Nevertheless, even with an adequate endometrial thickness, many embryos have been unable to implant. In order to delineate the factor that affected endometrial receptivity for women in this category, we only enrolled those who had an endometrial thickness ≥ 10 mm. The data available so far has revealed that the EPDA was an important factor that determined the probability of implantation. Women having a larger EPDA had a better chance of achieving successful implantation than those with a smaller EPDA.

An EPDA cutoff value of 5 mm^2 was utilized in this study. Women with poor endometrial vascularity (EPDA < 5 mm^2) demonstrated a significantly lower pregnancy rate and implantation rate than those who had adequate vascularity under the conditions of similar age, BMI, peak estradiol level and embryo number. Furthermore, the EPDA manifested as a unique parameter that was independent of endometrial thickness and uterine artery perfusion. The results confirmed our previous assumption that the uterine artery Doppler measurements were not representative of endometrial receptivity because of their basis on flow to the entire uterus.

Nevertheless, the predictive role of EPDA in embryo implantation was limited. Although containing an unfavorable EPDA, about 1 out of 4 women in this category achieved an intrauterine pregnancy. It may

be due to the fact that endometrial blood flow is not the sole factor affecting endometrial receptivity. Some other factors such as endometrial echo pattern, leukemia inhibitory factor (LIF) and transforming growth factor β (TGF β) might also play important roles.

In an IVF program, the appropriate number of embryos to be transferred remains a dilemma in that the higher number of transferred embryos results in not only a higher pregnancy rate but also in an increase in multiple pregnancies. In our study, after transferring similar numbers of embryos, the multiple pregnancy rate was about four times higher in women having cycles with an EPDA ≥ 5 mm^2 in comparison with those with an EPDA < 5 mm^2. Therefore, the number of transferred embryos might be determined on the basis of the quantity of intra-endometrial vascularity in addition to the endometrial thickness, so as to improve the reproductive outcome for those with poorly vascularized endometrium, and to reduce the potential risk of multiple pregnancies for those containing adequate intra-endometrial vascularity. For women with a small EPDA, cryopreservation might be suggested if a good freezing program is available. A prospective study is necessary to further verify this assumption.

Investigators had found various regimens such as low dose aspirin, heparin and nitroglycerin in improving endometrial receptivity due to their promoting effects on uterine perfusion. In our following course in the same patient, the EPDA values between the study cycle and the subsequent one were similar. Therefore, the endometrial vascularity seemed to be a persistent character. The measurement of EPDA might serve as an alternative method in re-evaluating the specific effects of these regimens on endometrium. Further large-scale studies are needed.

五、計畫結果自評

We accomplish this study on the basis of the plan that we presented previously. The results are clinically important. We will use it as a guideline in the future practices. The subsequent studies are also ongoing now.

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