Hysteroscopic appearance of the mid-secretory endometrium: relationship to early phase pregnancy outcome after implantation

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A total of 172 patients who underwent hysteroscopic assessment of the endometrium and then became pregnant, was analysed retrospectively to explore the relationship between endoscopic findings and early phase pregnancy outcome after implantation. Histological examination of the endometrium and assay of serum progesterone and oestrogen were carried out simultaneously with hysteroscopy. Of 172 patients, 12 were excluded. Of the remaining 160 patients, 62 (38.8%) were classified endoscopically as having ‘good’ mid-secretory endometrium and 98 (61.3%) as ‘poor’, between one and four cycles prior to the conception cycle. There were no clinical differences between these two groups, except that the frequency of patients with a history of early abortion was significantly higher in the ‘poor’ group (25.5%) than in the ‘good’ group (8.1%) (P < 0.05). Of 160 pregnancies, 118 persisted successfully to live birth, but 42 ended in early pregnancy loss. The incidence of early abortion was significantly higher in the ‘poor’ group (33.7%) than in the ‘good’ group (14.5%) (P < 0.05). Significant differences were observed between the two groups for histological dating of the endometrium (P < 0.05) but not for serum progesterone and oestradiol concentrations or progesterone:oestradiol ratio. In conclusion, our data suggest that the hysteroscopic appearance of the mid-secretory endometrium at this stage of the menstrual cycle is a better prognostic factor for pregnancy outcome than hormonal data.

Key words: hysteroscopy/mid-secretory endometrium/ miscarriage

Introduction

The endometrium of the secretory phase is morphologically and functionally well-prepared for implantation and the early development of the fertilized ovum. The maturation process of the endometrium is mediated by ovarian sex steroid hormones. In clinical practice, morphological information on endometrial maturation is available from histological evaluation and ultrasound assessment. Hysteroscopy is not yet used routinely to observe the morphological condition of the endometrium, although it has been employed in studying endometrial pathologies which might cause female infertility (De Jong et al., 1990; Nagele et al., 1996). Previous studies have shown that hysteroscopic observations could provide useful morphological information to evaluate the functional state of the endometrium, based on serial phase-specific changes in appearance of glandular openings and vasculature on the endometrial surface in subjects with normal ovulatory cycles (Inafuku, 1992; Sakamoto et al., 1992). These authors demonstrated that the assessments of the mid-secretory endometrium could be classified as ‘good’, characterized by ring-type glandular openings showing maximum glandular secretion and well-developed varicose-like vessels, or ‘poor’, characterized by dot- (no secretory) and/or punctate-type (early secretory activity) glandular openings and fine vasculature and that, in IVF and embryo transfer cycles, the pregnancy rate was significantly higher in patients with ‘good’ hysteroscopic findings than in patients with ‘poor’ findings. In this context, it was felt to be of interest whether or not the hysteroscopic appearance of the endometrium is associated with early pregnancy loss as well as with implantation failure of fertilized ovum.

The aims of the present study using hysteroscopic assessment of the mid-secretory phase endometrium were: (i) to explore whether or not there is a relationship between hysteroscopic findings and early phase pregnancy outcome after implantation; and (ii) to compare endoscopic findings with histological and endocrinological backgrounds.

Materials and methods

Patients

All 172 patients who had undergone hysteroscopic examination of the endometrium at our University Hospital, Okinawa, Japan, between January 1993 and December 1997, and became pregnant, were considered for the study. Patients with chemical or subclinical abortion, i.e. only elevation of human chorionic gonadotrophin (HCG) without clinical signs of pregnancy in IVF/embryo transfer cycles, were not included in the study. The vast majority of patients underwent outpatient hysteroscopy as part of the overall evaluation of infertility or repeated early abortion. At diagnosis of pregnancy, the mean age of the patients was 34.8 ± 3.8 (range 25–44) years. Of 172 pregnancies, 77 were spontaneous, while 95 were assisted by ovulation induction, IVF/embryo transfer and intrauterine insemination by the husband (IUI).

These pregnant patients were analysed retrospectively with special reference to a possible relationship between endoscopic findings and pregnancy outcome. Nine patients, whose pregnancies ended in ectopic pregnancy or in miscarriage suspected to be due to significant
Hysteroscopic view of endometrium and pregnancy outcome

Hysteroscopy and classification of findings

Hysteroscopy was scheduled in a menstrual cycle not manipulated with any hormonal agents. Patients were well informed regarding the procedure and were asked to prevent conception in the cycle. All patients ovulated and the procedure was carried out on days 7–9 after ovulation; basal body temperature (BBT) was monitored and/or the collapse of developing follicles documented by transvaginal ultrasound. The procedure was usually carried out without anaesthesia, but occasionally a paracervical block (PCB) of 10 ml 1% lidocaine was necessary. Lidocaine is not thought to have vaso-constrictive or dilative effects. No apparent change was observed in vasculature between pre- and post-PCB in those patients receiving PCB in the middle of hysteroscopy. Patients received cleansing and disinfection of their vulva and vagina in the dorsal lithotomy position. A rigid hystroscope which had a 4.5 mm outer diameter and gave a fo-oblque vision of 30° with the optical axis (Model A3726, Olympus, Tokyo, Japan) was inserted into the uterus transcervically and the uterine cavity was cleaned and expanded by irrigation with 5% glucose solution. After introducing the hystroscope through the internal uterine os, the uterine cavity was scanned thoroughly. After completion of the examination, endometrial biopsy was performed in 32 patients who agreed to the procedure.

Hysteroscopic findings were observed and diagnosed by more than three gynaecologists, including the authors (H.M. and K.K.) using videotape recordings. Endoscopic assessments of the mid-secretory endometrium on days 7–9 after ovulation were classified according to the appearance of both the glandular openings and the blood vessels on the endometrial surface as either ‘good’ when ring-type glandular openings and well-developed vascular networks were visualized over almost the whole endometrium (Figure 1a,b,c), or ‘poor’ when dot- and/or punctate-type glandular openings and fine vasculature were predominantly observed (Figure 2a,b,c; Sakumoto et al., 1992).

Histological dating of the endometrium

The specimens biopsied in 32 patients were submitted to conventional light microscopic examination to compare the observed hysteroscopic findings with histology of the endometrium. Post-ovulatory day of the cycle was specified by one of the authors (K.N.) according to Noyes’ criteria for dating the endometrium (Noyes et al., 1950; Noyes and Haman, 1953) with no clinical information. Histological assessments were classified as ‘in-phase’ endometrium when they were appropriate to within 2 days of the biopsy day and as ‘out-of-phase’ endometrium when they lagged >2 days behind the biopsy (Noyes and Haman, 1953; Davis et al., 1989).

Hormone assay

Serum progesterone and oestradiol, taken on the day of hysteroscopy, were measured in 83 patients by enzyme immunoassay kits (DPC Estradiol and Progesterone assay kits; Diagnostic Products Corporation, CA, USA). The normal values for progesterone and oestradiol in the luteal phase (days 3–15 after ovulation) were 0.2–31.6 ng/ml (SI conversion factor = 3.18) and 0.009–0.230 ng/ml (SI conversion factor = 3.671) respectively.

Ultrasonographic diagnostic criteria of blighted ovum and embryonic death

In this study, early pregnancy loss (early spontaneous abortion) was categorized into blighted ovum or intrauterine embryonic death, based on the findings of serial transvaginal ultrasonography. Blighted ovum was designated in cases where embryonic echoes were never visualized in the gestational sac, and embryonic death in cases where embryo heart action, which had been once identified, disappeared within 11 weeks of gestation (menstrual age).

Statistical analyses

Analysis of variance (ANOVA) and χ² or Fisher’s exact tests were used to assess the differences for clinical items, and histological and hormonal findings between the ‘good’ and ‘poor’ groups. P < 0.05 was considered to be statistically significant.

Results

Clinical profile of patients according to hysteroscopic findings

Of 160 patients, 62 (38.8%) were endoscopically diagnosed as having ‘good’ mid-secretory endometrium and 98 (61.3%) ‘poor’ mid-secretory endometrium, between one and four cycles prior to the conception cycle. The clinical characteristics of these two groups are shown in Table I. The mean age in the ‘poor’ group was almost equal to that in the ‘good’ group. The frequency of multi-gravid patients did not differ between the two groups, but the frequency of patients with a history of early abortion was significantly higher in the ‘poor’ group (25.5%) than in the ‘good’ group (8.1%) (P = 0.0059). There were no differences either in the frequency of patients with infertility factors or the distribution pattern of infertility factors between the two groups, and the overwhelming majority of patients had infertility factors.

Assisted intervention of index pregnancies

As shown in Table II, of the 160 pregnancies occurring after hysteroscopic examination, 68 (42.5%) were spontaneous with no assisted intervention. It was noted that 32 of these 68 pregnancies occurred in patients who had undergone hysterosalpingography or selective salpingography alone, several months prior to conception. Of the remaining 92 pregnancies, 11 were assisted by luteal support with HCG, 15 occurred after ovulation induction with clomiphene citrate or human menopausal gonadotrophin (HMG)–HCG, 61 after IVF/embryo transfer and five after IUI from the husband. There were no significant differences between the two groups for any of these assisted interventions.

Outcome of pregnancies according to hysteroscopic findings

Of the 160 pregnancies, 118 persisted successfully to live birth (102 term and 16 preterm), but 42 ended in early pregnancy loss (20 in embryo deaths and 22 in blighted ova) (Table II). No patients were encountered who suffered from late abortion at 12–21 weeks gestation. The incidence of early pregnancy loss was significantly higher in the ‘poor’ group (33.7%) than in the ‘good’ group (14.5%) (P = 0.0073). The incidence of embryonic death in these early spontaneous abortions was relatively higher in the ‘poor’ group (51.5%) compared with that in the ‘good’ group (33.3%), although the difference was not statistically significant.

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Figure 1. ‘Good’ hysteroscopic findings of the mid-secretory phase endometrium. (a) Ring-type glandular openings, and (b) well-developed and (c) engorged vascular networks can be seen over the whole endometrium. (d) The distended tortuous glands with intra-luminal wisps of secretion which were lined by low columnar to cuboidal epithelial cells, (e) opened widely through a protruded brim. (d) Scale bar = 25 μm; and (e) scale bar = 10 μm.

Histological and hormonal backgrounds of hysteroscopic appearance

Endometrial specimens from 32 patients were examined to specify the post-ovulatory day of the cycle according to histological criteria for dating the endometrium (Table III). In 20 patients in the ‘poor’ group, ‘in-phase’ endometrium was diagnosed in 13 (65.0%), and ‘out-of-phase’ endometrium was diagnosed in seven (35.0%). In the ‘good’ group, ‘in-phase’ endometrium was documented in all 12 patients examined. Thus, histological findings were significantly different between the two groups ($P = 0.0204$). In the tissue sections taken
Figure 2. ‘Poor’ hysteroscopic findings of the mid-secretory phase endometrium. (a) Dot- and punctate-typed glandular openings, and (b) sparse or (c) fine vasculature are observed. (d) The tortuous glands which were lined by columnar epithelial cells containing intracellular vacuoles, (e) opened with relatively narrow openings. (d) Scale bar = 25 µm; and (e) scale bar = 10 µm.

from the ‘good’-appearing endometrium, the distended tortuous glands with intra-luminal wisps of secretion, which were lined by low columnar to cuboidal epithelial cells, opened widely through a protruded brim, making a ring-type appearance on the endometrial surface (Figure 1d,e). On the other hand, in those from the ‘poor’-appearing sites of the endometrium, the tortuous glands, which were lined by columnar epithelial cells containing intracellular vacuoles, opened with relatively narrow openings, giving rise to a spot- or punctate-like appearance on the endometrial surface (Figure 2d,e).

Serum samples from 83 patients were submitted to progesterone and oestradiol assays (Table IV). There were no significant
Table I. Clinical profile of patients according to hysteroscopic findings of the mid-secretory phase endometrium. Data are presented as n with percentages shown in parentheses

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>‘Good’ (n = 62)</th>
<th>‘Poor’ (n = 98)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years, mean ± SD)</td>
<td>34.7 ± 4.5</td>
<td>35.0 ± 4.4</td>
<td>NSb</td>
</tr>
<tr>
<td>Primi-/multi-gravid</td>
<td>33 (53.2)/29 (46.8)</td>
<td>46 (46.9)/52 (53.1)</td>
<td>NSc</td>
</tr>
<tr>
<td>Cases with history of early abortion</td>
<td>5 (8.1)</td>
<td>25 (25.5)</td>
<td>0.0059d</td>
</tr>
<tr>
<td>Cases with infertility factors</td>
<td>60 (96.8)a</td>
<td>90 (91.8)</td>
<td>NSc</td>
</tr>
<tr>
<td>Male factor</td>
<td>15 (25.0)</td>
<td>17 (18.9)</td>
<td>NSc</td>
</tr>
<tr>
<td>Tubal pathology</td>
<td>30 (50.0)</td>
<td>46 (48.4)</td>
<td></td>
</tr>
<tr>
<td>Ovulatory dysfunction</td>
<td>16 (26.7)</td>
<td>25 (27.8)</td>
<td></td>
</tr>
<tr>
<td>Others</td>
<td>7 (11.7)</td>
<td>13 (14.4)</td>
<td></td>
</tr>
</tbody>
</table>

NS = not significant.
*aMultiple infertility factors were observed in five cases in the ‘good’ group and in seven in the ‘poor’ group.
*bAnalysis of variance.
*cχ² test.
*dFisher’s exact test.

Table II. Assisted intervention to and outcome of index pregnancies in two groups. Data are presented as n with percentages shown in parentheses

<table>
<thead>
<tr>
<th>Items</th>
<th>‘Good’ (n = 62)</th>
<th>‘Poor’ (n = 98)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Intervention</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No intervention</td>
<td>27 (43.5)</td>
<td>41 (41.8)</td>
<td>NSc</td>
</tr>
<tr>
<td>Luteal supporta</td>
<td>7 (11.3)</td>
<td>4 (4.1)</td>
<td></td>
</tr>
<tr>
<td>Ovulation inductionb</td>
<td>5 (8.1)</td>
<td>10 (10.2)</td>
<td></td>
</tr>
<tr>
<td>IVF/embryo transfer</td>
<td>22 (35.5)</td>
<td>39 (39.8)</td>
<td></td>
</tr>
<tr>
<td>IUI</td>
<td>1 (1.6)</td>
<td>4 (4.1)</td>
<td></td>
</tr>
<tr>
<td>Pregnancy outcome</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ongoing pregnancy</td>
<td>53 (85.5)</td>
<td>65 (66.3)</td>
<td>0.0073c</td>
</tr>
<tr>
<td>Early pregnancy loss</td>
<td>9 (14.5)</td>
<td>33 (33.7)</td>
<td></td>
</tr>
<tr>
<td>Details:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Embryo death</td>
<td>3 (33.3)</td>
<td>17 (51.5)</td>
<td>NSd</td>
</tr>
<tr>
<td>Blighted ovum</td>
<td>6 (66.7)</td>
<td>16 (48.5)</td>
<td></td>
</tr>
</tbody>
</table>

NS = not significant; IUI = intrauterine insemination by husband.
*aLuteal support in IVF/embryo transfer cycles is not included.
*bOvulation induction in IVF/embryo transfer cycles is not included.
*cχ² test.
*dFisher’s exact test.

Table III. Histological dating of the endometrium in two groups. Endometrial biopsies were taken on days 7–9 after ovulation. Data are presented as n with percentages shown in parentheses

<table>
<thead>
<tr>
<th>Dating⁴</th>
<th>‘Good’ (n = 12)</th>
<th>‘Poor’ (n = 20)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Appropriate</td>
<td>12 (100)</td>
<td>13 (65.0)</td>
<td>0.0204b</td>
</tr>
<tr>
<td>Lag of &gt;2 days</td>
<td>0</td>
<td>7 (35.0)</td>
<td></td>
</tr>
</tbody>
</table>

⁴Post-ovulatory day of the cycle was specified by histological criteria for dating the endometrium (Noyes et al., 1950).
*bFisher’s test.

differences for serum progesterone and oestradiol concentrations and progesterone:oestradiol ratio between ‘good’ and ‘poor’ groups.

Discussion

In 1992, the hysteroscopic appearance of the secretory endometrium was described (Sakumoto et al., 1992). Endoscopic findings were categorized into ‘good’ and ‘poor’ groups, by characterizing the glandular openings and the vascular networks on the endometrial surface. ‘Good’ appearance is of ring-type glandular openings, which show the maximum stage of gland secretion, with well-developed varicose-like vessel networks. Pregnancy rates of IVF/embryo transfer cycles were significantly higher in the ‘good’ group than in the ‘poor’, strongly suggesting that hysteroscopic findings of the mid-secretory endometrium were related to functional conditioning of the endometrium for implantation. It was also documented in our unpublished pilot study that there were no significant variations within individuals during at least five or six different menstrual cycles in the classification of their endometrial appearance. However, these hysteroscopic data were not compared with the histological and endocrinological backgrounds. In the present study, we examined whether the incidence of early pregnancy loss after implantation was significantly different between these two groups, as is the pregnancy rate in IVF/embryo transfer cycles, and whether there is a relationship between hysteroscopic, histological and hormonal findings.

First, of the 160 patients studied, 98 (61.3%) had been
hormone secretion by the corpus luteum, though the underlying
Bergh and Navot, 1992), is often a direct result of decreased
implantation of the fertilized ovum (Formigli
with day of the cycle. This delayed maturation, which cannot
implanted ovum. Appearance might not be well prepared for early development of the
result not from chromosomal abnormalities but from poor
abortions occurring at or before 8 weeks (Kajii
than chromosomal abnormality as a cause of early pregnancy
conditioning of the endometrium might be more important
was not statistically signi
appeared to be relatively higher in the ‘poor’ group (51.5%)
and/or with a history of early abortion, although the percentage of ‘poor’ appearance
is unknown in the fertile female population. Secondly, the
frequency of patients with a history of early spontaneous abortion was significantly higher in the ‘poor’ group than in the ‘good’ group. This suggests that unfavourable hysteroscopic findings are associated with early pregnancy loss after implantation. Thirdly, there were no significant differences between the two groups for any of the assisted interventions of index pregnancies. Therefore, it seems to be valid to analyse the outcome of pregnancies in association with hysteroscopic findings of the two groups. Thus, the incidence of early spontaneous abortion was significantly higher in the ‘poor’ group than in the ‘good’ group. The incidence of 14.5% in the ‘good’ group was similar to the abortion rate in the general population of women. It is also noteworthy that the incidence of intrauterine embryo death in early spontaneous abortion appeared to be relatively higher in the ‘poor’ group (51.5%) than in the ‘good’ group (33.3%), although the difference was not statistically significant. In the ‘poor’ group, poor conditioning of the endometrium might be more important than chromosomal abnormality as a cause of early pregnancy loss. The high prevalence of chromosomal abnormality in abortions occurring at or before 8 weeks (Kajii et al., 1980) suggests that the majority of blighted ova could result from lethal chromosomal abnormalities. On the other hand, the low abortion rate after ultrasound-proved viability of embryo/fetus in early pregnancy (Simpson et al., 1987; Rosen et al., 1990) appears to suggest that the majority of embryo deaths might result not from chromosomal abnormalities but from poor conditioning of the endometrium. Thus, the mid-secretory endometrium diagnosed to be endoscopically ‘poor’ in appearance might not be well prepared for early development of the implanted ovum.

Luteal phase defect is defined as a lag of >2 days in histological maturation of the secretory endometrium compared with day of the cycle. This delayed maturation, which cannot allow for the timely opening of the temporal window for implantation of the fertilized ovum (Formigli et al., 1988; Bergh and Navot, 1992), is often a direct result of decreased hormone secretion by the corpus luteum, though the underlying causes may be multiple. One of the controversies concerning the histological diagnosis of luteal phase defect has revolved around issues of technique and timing of the endometrial biopsy (Li et al., 1989; Castelbaum et al., 1994). In the present study, where hysteroscopy-oriented biopsies were performed, ‘out-of-phase’ secretory endometrium was observed in 35.0% of the ‘poor’ group, but in none of the ‘good’ group, although the size of samples examined was small. In other words, more than one-third of the endometrial samples with unfavourable hysteroscopic findings demonstrated delayed histological maturation compared with day of the cycle. This suggests that hysteroscopic appearance of the secretory endometrium may reflect the histological endometrial maturation status. It has been noted by several authors (Vanrell and Balasch, 1986; Tulppala et al., 1991; Peters et al., 1992) that the incidence of luteal phase defect may be higher in patients with a history of recurrent abortion. Thus, it is again conceivable that the mid-secretory endometrium with unfavourable hysteroscopic appearance might be inadequately conditioned for early development of the implanted ovum. There have been a number of morphological, immunohistochemical and molecular–biological observations on cyclical changes of the endometrial glands and stroma. In particular, immunohistochemical profiles of oestrogen and progesterone receptors, tumour-associated glycoprotein 72, placental protein 14, and carbohydrate in glandular epithelial cells have been demonstrated to be associated with secretory differentiation of glandular function (Waites et al., 1988; Ravn et al., 1992; Osteen et al., 1992). Comparative studies between hysteroscopic appearance and these observations of the endometrium should be performed in the near future.

Hormonal data in luteal phase defect have been in some dispute to date. Low FSH concentrations and low FSH:LH ratios in the follicular phase, low serum concentrations of progesterone and oestrogen in the luteal phase, and decreased response of the endometrium to progesterone have been implicated (Soules et al., 1989). In clinical practice, attention has been directed to measurement of serum progesterone concentrations for diagnosing luteal phase defects (Cumming et al., 1985). Most important, however, is the impressive evidence documenting the lack of a relationship between serum progesterone concentrations and endometrial histology. This is also the case in our study which examined a possible correlation between serum progesterone concentrations and hysteroscopic findings. Furthermore, no significant association

<table>
<thead>
<tr>
<th>Table IV. Progesterone, oestradiol concentrations and progesterone:oestradiol ratio in serum samples from the two groups. Serum samples were taken on days 7–9 after ovulation. Data are presented as mean ± SD with range shown in parentheses</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Hormone</strong></td>
</tr>
<tr>
<td>Progesterone (ng/ml)</td>
</tr>
<tr>
<td>Oestradiol (ng/ml)</td>
</tr>
<tr>
<td>Progesterone:oestradiol ratio</td>
</tr>
</tbody>
</table>

NS = not significant.
SI conversion factors are 3.18 for progesterone and 6.671 for oestradiol.
*aAnalysis of variance.

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was observed between luteal progesterone:oestradiol ratios and hysteroscopic appearance in the present study, although it has been reported that, after ovulation induction, a high luteal progesterone:oestradiol ratio is associated with ongoing pregnancies and a lower progesterone:oestradiol ratio is seen in cycles leading to spontaneous abortion (Maclin et al., 1990).

In conclusion, our data suggest that the hysteroscopic appearance of the mid-secretory endometrium is a better prognostic factor for pregnancy outcome than any hormonal data investigated at this stage of the menstrual cycle.

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References

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