Clinical Application of Ultrasound in Infertility: From Two-dimensional to Three-dimensional

Chun-Kai Chen*, Hsien-Ming Wu, Yung-Kuei Soong

Transvaginal probes have enhanced sonographic depiction of the uterus and ovaries over that obtained with conventional transabdominal scans. Cyclic variations of female pelvic hemodynamics and angiogenesis can be studied with Doppler ultrasound. The power Doppler has a threefold increase in sensitivity compared with conventional color Doppler imaging at detecting low velocity flow. There have been numerous investigations on the applications of the assessment of uterine and perifollicular vascularity and their possible relation with the treatment of infertility. However, the main applications in infertility so far are follicular monitoring and endometrium measurement by traditional two-dimensional (2-D) ultrasound. Nowadays, sonography plays a vital role in tracking follicular development and endometrial assessment in patients receiving ovulation-induction medication. The sonographic information can be coupled with estradiol values to provide an accurate assessment of the presence or absence and number of mature follicles. The maximal follicle size is the main point to consider giving human chorionic gonadotropin and ovum pick-up.

Three-dimensional (3-D) ultrasound is a new imaging modality which is being introduced into clinical practice. It has been proved that 3-D ultrasound is a very highly reproducible technique. With 3-D ultrasound, a volume of a region of interest can be acquired and stored. This volume can be further analyzed in several ways, such as navigation, multiplanar display and surface rendering or volume calculation. Power Doppler ultrasound, in combination with 3-D ultrasound, allows for a whole assessment of relevant vessels and quantitative assessment of vessel density and perfusion within a specified area. A whole evaluation is then possible for endometrial and subendometrial vascularization and also for ovarian stromal vascularity. Further, structure anomaly, such as septate uterus, can be understood more clearly with 3-D ultrasound. However, data in the literature on 3-D ultrasound assessment of endometrial receptivity to predict in vitro fertilization (IVF) outcome are controversial. Ovarian volume and antral follicle numbers have been shown to be an indicator of ovarian reserve and reproductive potential. A recent comparative meta-analysis has demonstrated that the predictive performance of ovarian volume toward poor response is clearly inferior compared with that of antral follicle count (AFC). AFC may be considered the test of first choice when estimating quantitative ovarian reserve before IVF. In combination with 3-D ultrasound, the use of power Doppler in examining ovarian stromal blood supply still fails to prove its role in predicting ovarian response and pregnancy in the IVF treatment.
and in the intrauterine insemination treatment. Although 3-D ultrasound has probably not replaced 2-D ultrasound, it is being increasingly used in the clinical setting. 3-D ultrasound has become an indispensable and auxiliary tool, alongside with 2-D ultrasound.

**KEY WORDS** — 2-D ultrasound, 3-D ultrasound, endometrium, ovarian follicles, power Doppler

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**Introduction**

The development and clinical application of transvaginal probes have greatly enhanced sonographic depiction of the uterus and ovaries over that obtained with conventional transabdominal scans. The major factors that have contributed to this enhancement include a shorter probe-to-target distance, which allows use of higher-frequency transducers, and the use of multi-element phased linear arrays, which afford more tightly focused beams with higher line densities than conventional real-time probes. Ultrasound has actually become one of the indispensable diagnostic tools in the field of obstetrics and gynecology. This review will focus on the clinical application in infertility only.

**Application of Traditional Transvaginal Sonography**

**Endometrium measurement [1,2]**

Transvaginal sonography allows for a more complete evaluation of the endometrium than conventional transabdominal techniques. It delineates specifically the endometrium with anterior-posterior (AP) length on long-axial scan and width on semi-axial scan. The sonographer must be careful to image the endometrium in the long- and semi-axial planes in which the two layers of the endometrium appear symmetric.

After menarche, the endometrium undergoes cyclic changes in its thickness and texture that can be related to the relative amount of serum estrogen and progesterone. During the menstrual phase, the endometrium appears as an echogenic interrupted interface of 1–4 mm in AP width. A hypoechoic outer layer can be identified surrounding the endometrium, corresponding to the inner layer of myometrium. This hypoechoic appearance of the inner myometrium may be related to the more compact configuration of the myometrial cells in this area or the relatively smaller content of connective tissue and smooth muscle of the intermediate and external layers of the myometrium. One to 2 mm vessels can be seen in the external myometrium representing a venous plexus in the outer portion of the myometrium.

During the proliferative phase, the endometrium thickens between 4 and 8 mm and has an isoechoic or slightly hyperechoic texture relative to the outer myometrium. In the late proliferative or periovulatory phase of endometrial development, a multilayered endometrium can be depicted. The inner hypoechoic area probably represents edema in the compact layer of the endometrium. As imaged in a semi-axial or semi-coronal plane, the endometrium has the configuration of a theta with respect to the hypoechoic areas. This finding has been described as an indication that ovulation is near. However, with transvaginal sonography, this finding is noted both prior to and immediately following ovulation [2]. In the secretory phase, the endometrium achieves a width of between 8 and 16 mm and is echogenic, most likely related to the increased mucus and glycogen within the glands as well as the increased number of interfaces created by tortuous glands in this phase. The endometrium typically achieves its greatest thickness in the midsecretory phase of a spontaneous cycle, measuring up to 14 mm in width.
The data in the Table serves as a guideline for normal values of endometrial thickness in women with cyclic menstruation [1].

**Follicular monitoring [1,2]**

When menarche begins, approximately 200,000 follicles remain per ovary. During the childbearing years, approximately 200 oocytes will be ovulated. This indicates that approximately 99.9% of primary oocytes become atretic or do not develop at all [2].

Maturation of the oocyte and follicle is responsive primarily to changes in follicle-stimulating hormone (FSH), luteinizing hormone (LH) and circulating levels of estradiol (E2). With the elaboration and release of FSH in the late secretory phase, there is a recruitment of a group of preantral follicles and development in a subsequent cycle. When the dominant follicle is selected, LH reinitiates meiosis of the oocyte and ovulation typically occurs within 36 hours of its “surge” in circulating levels. E2 is synthesized by the granulosa cells and provides important feedback to the pituitary in the production of FSH and LH.

Sonography can track the developing follicles, beginning at the time they measure between 3 and 5 mm. In the spontaneous cycle, there are usually one or at the most two follicles that develop to approximately 10 mm in size. As the follicle matures, more and more fluid is elaborated into its center and the number of granulosa cells lining the inside wall of the follicle increases. The oocyte itself, which is less than 0.1 mm, is surrounded by a cluster of granulosa cells. This complex is termed the cumulus oophorus. It measures approximately 1 mm and can be depicting occasionally inside mature follicles.

As the follicle reaches maturity, its inner dimensions range from 17 to 25 mm. Intrafollicular echoes may be observed with mature follicles, probably rising from clusters of granulosa cells that shear the wall close to the time of ovulation. After ovulation, the follicular wall becomes irregular as the follicle becomes “deflated”. The corpus luteum usually appears as an echogenic structure with a small hypoechoic center. As the corpus luteum develops some 4–8 days after ovulation, it appears as an echogenic structure approximately 15 mm in size. In addition, the undesirable development of multiple immature follicles, rather than development of a single dominant follicle, can be recognized in patients with polycystic ovaries. Sonographic finding has been included as one of the three criteria for polycystic ovary syndrome [3,4]. Presence of 12 or more follicles in each ovary measuring 2–9 mm in diameter and/or increased ovarian volume (>10 mL) fulfill the sonographic findings.

Nowadays, sonography plays a vital role in tracking follicular development in patients receiving ovulation-induction medications. Since the maturity of the oocyte can only be indirectly inferred by the size of the follicle, the sonographic information can be coupled with E2 values to provide an accurate assessment of the presence or absence and number of mature follicles. The maximal follicle size is the main point to consider giving human chorionic gonadotropin and ovum pick-up.

In addition to delineation of changes in follicle size and morphology, sonography can depict the presence of intraperitoneal fluid. It is not uncommon to have approximately 1–3 mL in the cul-de-sac prior to ovulation. When ovulation occurs, there is typically between 4 and 5 mL within the cul-de-sac. When the patient is scanned with a fully distended bladder, the fluid may be located outside of the cul-de-sac, surrounding bowel loops in the lower abdomen and upper pelvis [2].

Measuring the volume of both ovaries or counting the number of small antral follicles by ultrasound has been suggested to predict well the ovarian reserve and outcomes of assisted reproductive technologies (ART) [5,6]. Till now, antral follicle count

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<th>Table. Transvaginal sonography of endometrial width: anterior-posterior (AP) dimension</th>
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<td><strong>Range (mm)</strong></td>
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<tr>
<td>Menstrual phase</td>
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<td>Proliferative phase</td>
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<td>Secretory phase</td>
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*Up to 10 mm if taking estrogen.
(AFC) is still considered the test of first choice to estimate quantitative ovarian reserve before in vitro fertilization (IVF) [7].

**Assessment of Neovascularization with Color Doppler and Power Doppler**

It has been suggested that neovascularization may be of prime importance in the growth and selection of ovulatory follicles. Several studies have examined the cyclic variations of female pelvic hemodynamics and angiogenesis using Doppler ultrasound [8,9]. Since then, transvaginal pulse Doppler has been used quite extensively to assess uterine and ovarian blood flow patterns in ART cycles [10–12]. Later, there have been increasing publications suggesting that the use of color flow imaging may assist in the management of ART cycles [13–15]. However, power Doppler or color angio-ultrasonography has aroused attention since its introduction. The power Doppler has a threefold increase in sensitivity compared with conventional color Doppler imaging at detecting low velocity flow [16,17]. There have been reports about applications in relation to the assessment of uterine [18] and perifollicular vascularity [19,20] and outcome of IVF. Similar applications were reported of perifollicular vascularity [21] and uterine perfusion and outcome of intrauterine insemination (IUI) [22,23].

**The Era of Three-dimensional (3-D) Ultrasound**

The advent of 3-D ultrasound has widened the field of clinical application. With this technology, any desired plane through an organ can be obtained. With 3-D ultrasound, a volume of a region of interest can be acquired and stored. This volume can be further analyzed in several ways, such as navigation, multiplanar display and surface rendering or volume calculation. Power Doppler ultrasound, in combination with 3-D ultrasound, allows for a whole assessment of relevant vessels and quantitative assessment of vessel density and perfusion within a specified area (Fig.).

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**Fig.** Example of three-dimensional (3-D) ultrasound for endometrial measurement in multiplanar image by the power Doppler mode. Transverse, longitudinal and frontal view in upper right, upper left and lower left plane, respectively. The rendering 3-D projection of the vascular images is shown in the lower right plane.
A whole evaluation is then possible for endometrial and subendometrial vascularization [24–26] and also for ovarian stromal vascularity [27–29].

With 3-D ultrasound, researchers focus on improvement of clinical application over two-dimensional (2-D) ultrasound. Endometrial volume measurement is a new tool, at least as good as endometrial thickness measurement in predicting pituitary suppression during IVF [30]. Endometrial and subendometrial blood flow measurement has been thought as indicators for uterine receptivity and outcome of treatment [31,32]. However, literature about 3-D ultrasound assessment of endometrial receptivity was reviewed by Alcazar [26]. He has found that data from studies analyzing the role of 3-D ultrasound for predicting IVF outcome are controversial. His explanation for these controversial findings was that it might be the different design of reported studies, especially the timing of ultrasound evaluation. Interestingly, endometrial volume in spontaneous cycles grows during follicular phase and remains constant through the luteal phase [33,34]. Endometrial vascularization increases during follicular phase, peaking 2–3 days before ovulation, decreasing thereafter and increasing again during mid and late luteal phase [35,36].

A variety of hormonal markers have been used to predict ovarian reserve in infertile women, such as cycle day 3 FSH, E2 and inhibin B, as well as challenge tests [37–40]. Ovarian volume and antral follicle numbers have been shown to be indicators of reproductive potential [5,6,41,42]. Ovarian volume has been claimed as a better predictor of ovarian reserve than several other predictors, including the AFC [43]. A meta-analysis showed that AFC performed better in the prediction of outcome in IVF than basal FSH [44]. Moreover, a recent comparative meta-analysis has demonstrated that the predictive performance of ovarian volume toward poor response is clearly inferior compared with that of AFC. AFC may be considered the test of first choice when estimating quantitative ovarian reserve before IVF. For the prediction of cases with a very low chance for pregnancy, ovarian reserve testing with the use of ultrasound appears inadequate [7].

As to the role of ovarian stromal vascularity detected by 3-D power Doppler flow indices, recent studies do not support it as a good predictor of ovarian response and pregnancy in the IVF treatment [29].

It has been postulated that increased ovarian stromal vascularity may lead to a greater delivery of gonadotropins to the granulosa cells of the developing follicles. Ovarian stromal vascularity can be assessed by color or power Doppler ultrasound. Power Doppler is better applied to the study of ovarian stromal vascularity, as it is more sensitive to lower velocities and essentially angle-independent [45]. Ovarian stromal vascularity has been measured by 2-D power Doppler ultrasound and there was no predictive value for the ovarian response [46]. In combination with 3-D ultrasound, the use of power Doppler in examining ovarian stromal blood supply still fails to prove its role in predicting ovarian response and pregnancy in IVF treatment [29] and IUI treatment [47].

Structure Anomaly

By using transvaginal ultrasound, it is possible to perform a precise assessment of the uterine morphology, including the endometrial lining and the outer shape of the uterine muscle. With instillation of isotonic saline solution into uterine cavity through a catheter, the intrauterine pathologies, such as submucous myomas, endometrial polyps and septate uteri, can be accurately diagnosed. The patency of bilateral tubes can be detected too. The procedure is called sonohysterography, which is particularly useful in infertile patients [48–50]. The color Doppler technique allows simultaneous visualization of the morphology and the vascular network, giving full information on the type of anomaly and the extent of the defect. The visualization of the myometrial portion is further enhanced by detection of myometrial vessels by the color Doppler technique. Furthermore, Doppler imaging can detect deficient intraseptal vascularity and/or inadequate endometrial development in patients with a septate uterus [51].
3-D ultrasound enables precise evaluation of the fundal indentation and the length of the septum in the case of septate uteri. However, the technique may give an incorrect impression of an arcuate uterus in patients with fundal location of a leiomyoma. Furthermore, shadowing caused by the uterine fibroids, irregular endometrial lining and decreased volume of the uterine cavity (in cases of intrauterine adhesions) are limitations of 3-D ultrasound. 3-D power Doppler has been used to detect the vascularization of uterine septa in a combined angiographic and gray-rendering mode. This approach allows simultaneous analysis of the morphology, texture and vascularization [51].

Conclusion

3-D ultrasound is a new imaging modality which is being introduced into clinical practice. It has been proved that 3-D ultrasound is a very highly reproducible technique. For the clinical stage of infertility, however, the main actor still remains as the traditional 2-D ultrasound. It seems that the chief applications in infertility, so far, are follicular monitoring and endometrium measurement. Although 3-D ultrasound has probably not replaced 2-D ultrasound, it is being increasingly used in the clinical setting. 3-D ultrasound has become an indispensable and auxiliary tool, alongside with 2-D ultrasound.

References


