Follicle Monitoring and Endometrial Correlation

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ABSTRACT

Thirty-percent of female infertility is due to ovulatory dysfunctions. Today ultrasound (transvaginal ultrasound scan + color + 3D) enables us to understand the follicular dynamics during normal and stimulated cycles. In this review paper, we have analyzed folliculogenesis, follicular monitoring, and measuring and have correlated clinically and presented simply the importance of 2D, 3D, color, volume, vocal, and endometrial perfusion.

Keywords: Endometrial scoring, Follicle, Follicle monitoring, Ovulation induction.

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INTRODUCTION

Folliculometry is the gold standard investigation to monitor follicular development and growth, check for signs of ovulation corpus luteum (CL) integrity, and endometrial growth and character. Ultrasound monitoring of follicular growth was first introduced in 1978 by Hackelöer and Robinson.¹ Transvaginal ultrasound scan (TVS) is now the norm for routine monitoring of follicular growth whether it may be a natural cycle, ovulation induction program, or controlled ovarian hyperstimulation (COH) in assisted reproduction technology (ART) cycles. Recently, color Doppler and 3D power Doppler (PD) parameters have been found to have added value in follicular monitoring. Color Doppler and PD provide qualitative and quantitative information of perfusion to the ovaries.²⁻⁴

Monitoring a Natural Cycle (Figs 1 to 4)

- Baseline scan is done on day 2 or 3 (Fig. 1).
- First follow-up scan can be done either on day 9 or 10 of the menstrual cycle (MC).
- The scans are repeated every 48 hours till the follicles reach 14 mm and then repeated every day.
- Follicle can rupture at any time once the follicle becomes more than 16 mm (Fig. 4).

Features of Ovulation on Ultrasonography (USG) (Fig. 5)

- Diminution in the follicle size or sudden collapse of the follicle
- Blurring of the follicle borders, which become crenated
- Appearance of intrafollicular echoes, which are more isoechogenic with respect to surrounding ovary
- Presence of a small amount of free fluid in the pouch of Douglas (POD)
- Thereafter, an irregular, slightly cystic structure representing the corpus luteum forms and slowly shrinks throughout the luteal phase of the cycle until luteolysis occurs before menses.

Follicular Growth in a Natural Cycle

- Small antral follicles (2–5 mm in diameter) appear in the ovary early in cohorts in the proliferative phase. (Wave theory of follicular recruitment.)
- The physiological decline in follicle stimulating hormone (FSH) level in the late follicular phase allows the selection of the single most sensitive follicle to continue to grow.

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- Further growth of the follicles occurs in relation to rising levels of FSH.
- The follicle, which has developed maximum receptors for FSH and luteinizing hormone (LH) will continue to grow, while the other follicles will undergo apoptosis and atresia (Fig. 3). Concept of FSH threshold and FSH window. An optimum threshold level of FSH is needed to stimulate follicular growth.
- Once the leading follicle reaches a diameter of approximately 14 mm, the daily growth rate is between 1.5 mm and 2.0 mm until a diameter of 22–25 mm is reached when ovulation occurs (Fig. 6).

Monitoring of an Ovulation Induction Cycle with Oral Agent

- A baseline scan is done always on day 2 or 3. Should be done before the start of any ovulation induction therapy.
- Ovulation induction drugs are initiated within 3 days of the MC if the follicular size is <10 mm, there are no ovarian cysts, the endometrial thickness is <5 mm, estradiol levels are <50 pg/mL, and the progesterone level is <1.5 ng/mL.
- Transvaginal ultrasound scan is usually performed 4–5 days after the last dose of the oral ovulation agent and then, every other day till the follicle is 14 mm, and then daily until a follicle of approximately 18–20 mm in diameter is seen.
- The ovulation trigger is given at follicle size ≥ 18 .

Figs. 1A and B: Antral follicle count

Fig. 2: Day 6–7 follicle stimulated ovary

Fig. 3: Dominant follicle

Monitoring for Ovulation Induction with Gonadotropins

- The gonadotropins are started on day 2 or 3.
- The first scan after the start of treatment of gonadotropins is done on the 4th day of the drug (day 7 of MC).
- Further adjustment of the gonadotropin dose and/or administration of gonadotropin-releasing hormone (GnRH) antagonist depends on serial USG findings and E2 levels on day 7 of MC or 3–4 days of gonadotrophin treatment.

A Rough Guide is as Follows

- At each scan, the size of all follicles documented, daily scans are done and response is charted on the ovulation (follicle) monitoring chart (Fig. 7) once follicular diameter 12 mm or more.

Figs 4A and B: Periovulatory follicle

Figs 5A and B: Signs of ovulation

If on day 4	If on day 7
<ul style="list-style-type: none"> Number of follicles <4, dose is increased by 37.5/75 IU. 	<ul style="list-style-type: none"> Rate of growth <2 to 3 mm/day and number of follicles <4 which are <12 mm in size dose increased by 37.5/75 IU.
<ul style="list-style-type: none"> Number of follicles 8, dose reduced by 37.5/75 IU. 	<ul style="list-style-type: none"> Rate of growth 2 to 3 mm/day and number of follicles >10, which are >12 mm in size the dose is decreased by 37.5/75 IU.

Administration of Trigger

The general principle of human chorionic gonadotropin (hCG)/antagonist trigger administration is as follows:

- In natural cycles and clomiphene citrate stimulated cycles—when the dominant follicle is 20–24 mm.
- In gonadotropin-stimulated cycles and intrauterine insemination (IUI)—when the dominant follicle is larger than 18–20 mm.
- In *in vitro* fertilization (IVF) cycles—when three follicles above 16–18 mm are present with good color flow. The follicle monitoring chart (Fig. 8) is plotted.

Fig. 6: Gonadotropin stimulated ovary

Color Doppler Studies of Ovarian Circulation

The perifollicular microvascular network is an essential element for the initiation and maintenance of follicular growth.^{5,6}

OVULATION MONITORING																																	
Fertile Days																																	
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Follicle Monitoring Cycle																																	
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Fig. 7: Follicle monitoring chart

Grading of perfollicular blood flow (PFBF): Based on the percentage of blood flow (BF) of the circumference of the follicle:

Grade I: BF <25% (Fig. 9).

Grade II: BF ≥25% but <50% (Fig. 10).

Grade III: BF ≥50% but <75%.

Grade IV: BF ≥75% (Fig. 11).

Clinical Application of Color and Pulse Doppler Parameters of Ovarian Follicles

- Studies have shown that Doppler parameters of follicles that have >75% (Fig. 11) of their surface perfused, ovarian stromal peak systolic velocity (PSV) of >10 cm/second, and resistance index (RI) of <0.4 to 0.48 contain mature oocytes of satisfactory quality and result in a better grade of embryos.^{7,8}
- Follicles having a PFBF of >50% (Fig. 10) have increased oocyte retrieval rate with more number of mature oocytes with a high fertilization rate and lower triploidy rates (Fig. 12).
- Rising PSV with steady low RI suggests that the follicle is close to rupturing (follicular PSV goes as high as 45 cm/second an hour before ovulation).
- On the contrary, steady or decreasing PSV with rising RI suggests the possibility of a luteinized unruptured follicle (LUF).
- It was also observed that the fertilization of a follicle with PSV of <10 cm/second has high chances of the embryo being chromosomally abnormal^{5,9,10} (Fig. 13).

- Doppler in the secretory phase gives an idea about the function of CL. Usually, the RI of the CL is between 0.35 and 0.50. In luteal phase deficiency (LPD), RI is 0.58 ± 0.04 , pulsatility index (PI) is 0.70 to 0.80, and PSV is between 10 and 15.

3D Volume and Doppler Parameters

- The follicular volume of 3–7 cm³ is optimum.^{11,12}
- Follicles without visualization of cumulus in all three planes are not likely to contain mature oocytes. The appearance of the intrafollicular cumulus structures by 3D USG has a good correlation with the recovery rate of the mature oocytes (Fig. 11).¹³
- The follicular volume between 3 and 7 cm³, presence of cumulus, the perfollicular vascular index (VI) between 6 and 20, and perfollicular flow index (FI) > 35 are associated with a better pregnancy rate (Fig. 14).^{11,12}
- The best predictors of IVF outcome are the ovarian FI using 3D ultrasound and power Doppler angiography (PDA) on the hCG day and the transfer of grade I embryos.⁶
- The assessment of multiple follicles is better done by sonography-based automated volume count (sonoAVC) (Figs 13 and 14).

Endometrium

Endometrial receptivity is pivotal for successful implantation and hence for conception. Therefore, in addition to ovarian follicles, endometrium also should be assessed for its morphological and functional characteristics (Figs 14 to 20).

Fig. 8: Ovarian and follicle volume (3D)

Fig. 9: 25% color flow in follicle (grade I)

Fig. 10: 50% color flow in follicle wall (grade II)

Fig. 11: 3D color flow follicle (grades III-IV)

Figs 12A and B: Color Doppler follicular flows

Endometrial Thickness

Endometrial thickness is measured as the sum thickness of the two opposing endometrial layers in the mid-sagittal plane. Few pregnancies have been noted in gonadotropin-induced

IUI cycles when the endometrium measured <7 mm on the day of hCG-induced ovulation.¹⁴ However, in several studies, optimum endometrial thickness has been found out to be 8–14 mm.^{15–20}

Fig. 13: Follicular volume

Figs 14A and B: 3D follicle volume

Fig. 15: Sonography-based automated volume count (sonoAVC)

Morphology

Morphologically, the endometrium has been graded as follows.

Grade I (best)—when it is a triple-line endometrium with the intervening area is as hypoechoic as the anterior myometrium. Grade II (intermediate)—when it is multilayered or triple line with the hypoechoic intervening area. Grade III (most unfavorable)—when it is a homogeneous isoechoic endometrium.

Breach or irregularity of endomyometrial junction is an indication of unhealthy endometrium and therefore poor receptivity.

Uterine Artery Doppler

Several authors have shown that the optimum uterine receptivity was obtained when the average pulsatility index of the uterine

artery was between 2 and 3 and RI should be <0.9 on the day of transfer or the day of hCG.²¹⁻²³

Endometrial and Subendometrial Vascularity

There are three zones of vascularity according to Applebaum:

Zone 1: when the vascularity on PD is seen only at endometriomyometrium junction; Zone 2: when vessels penetrate through the hyperechogenic endometrial edge; Zone 3: when it reaches the intervening hypoechoic zone; and Zone 4 when they reach the endometrial cavity.

Vascularity in zones 3 and 4 represents a mature endometrium and is correlated to have better implantation rates.

Endometrial Volume

The endometrial volume by 3D USG having an optimum pregnancy rate has been described as 2–13 mL. The calculation of endometrial volume is particularly useful in cases of synechiae, adenomyosis, and uterine anomalies to predict the outcome of treatment. Endometrial and subendometrial volume increase rapidly during the follicular phase and then remain almost unchanged during the luteal phase.

Endometrial Vascularization Using 3D Power Doppler

Endometrial vascularization is calculated by measuring the VI, FI, vascular flow index (VFI), and flow vessel quotient. Higher subendometrial Doppler indices, i.e., VI (>1), FI (>31), and VFI (>0.25) on the day of hCG have been found to have a higher conception rate.²⁴⁻²⁶

CONCLUSION

Folliculometry is the cornerstone of an infertility management program. Ultrasound can alone be used to accurately monitor ovulation index (OI) therapy for both *in vivo* and IVF by successfully measuring endometrial thickness and size of ovarian follicles and correlates strongly with serum estradiol concentrations. Color

Fig. 16: Endometrial patterns

Figs 17A and B: UT. A. In a menstrual cycle

Doppler and 3D PD parameters can have an adjunctive role to usual folliculometry. The follicular and endometrial physiological status can be better understood with these newer parameters.

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Fig. 18: Spiral artery perfusion

Fig.19: Endometrial volume

Figs 20A to C: Endometrial perfusion

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