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REVIEW ARTICLE

Follicular Monitoring

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ABSTRACT

Assessment of the follicular maturity and endometrial receptivity and the time of hCG is one of the key factors for success of all ART procedures. Maturation of the follicle and the endometrium, ovulation and leutinization is a process of multiple biochemical, morphological and vascular changes. The vascular changes are reflection of the biochemical changes and can be studied by color Doppler. 3D ultrasound gives a better assessment of the follicular and endometrial size, that is the anatomical maturity, than 2D ultrasound and 3D power Doppler gives not only qualitative but also quantitative idea of global vascularity, that is the reflection of functional/physiological maturity. Follicular vascularity distribution and flow indices can be better parameters of follicular quality and can be more reliable parameters to decide the time of hCG and IUI. Endometrial assessment can be more meaningful if its morphology is studied more in detail along with abundance of its vascularity as well as flow indices. Thus, deciding correct time of hCG can improve conception rates in ART cycles.

Keywords: pre-hCG scan, Follicular maturity, Endometrial receptivity, 3D power Doppler.

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INTRODUCTION

The advances in ultrasound (US) technology have changed the management of infertility. With the advent of color Doppler, pulse Doppler, three-dimensional (3D) US and 3D power Doppler, the previously unexplained causes of failure of fertilization and implantation can now be explained. 3D and 3D power Doppler can now be used for the assessment of the maturity of the follicle and receptivity of the endometrium and can give better idea about the functional maturity of the follicle and the endometrium, to decide the timing of human chorionic gonadotropin (hCG) administration for all assisted reproductive techniques for better pregnancy rates. The accuracy of diagnosis and monitoring of infertility treatments, such as ovulation induction, has greatly increased because of the availability of sophisticated US technology and equipment.[1]

Assessment of the follicular maturity and endometrial receptivity and the time of hCG is one of the key factors for success of all ART procedures. The follicular maturity can be assessed by estradiol levels, but frequent assessment of blood estradiol level is a cumbersome procedure. Since, the advent of the transvaginal ultrasound this has been a preferred method for assessment of follicle and endometrium.

Earlier, the follicular size of 16 to 18 mm and the endometrial thickness of 8 mm were considered optimum parameters for hCG administration for ovulation trigger. But this is assessing only the anatomical maturity of follicle and endometrium. Maturation of the follicle and the endometrium, ovulation and leutinization is a process of multiple biochemical, morphological and vascular changes. The vascular changes are reflection of the biochemical changes and can be studied by color Doppler. 3D US gives a better assessment of the follicular and endometrial size, that is the anatomical maturity, than two-dimensional (2D) US and 3D power Doppler gives not only qualitative but also quantitative idea of global vascularity, that is the reflection of functional/physiological maturity.

Follicular Assessment

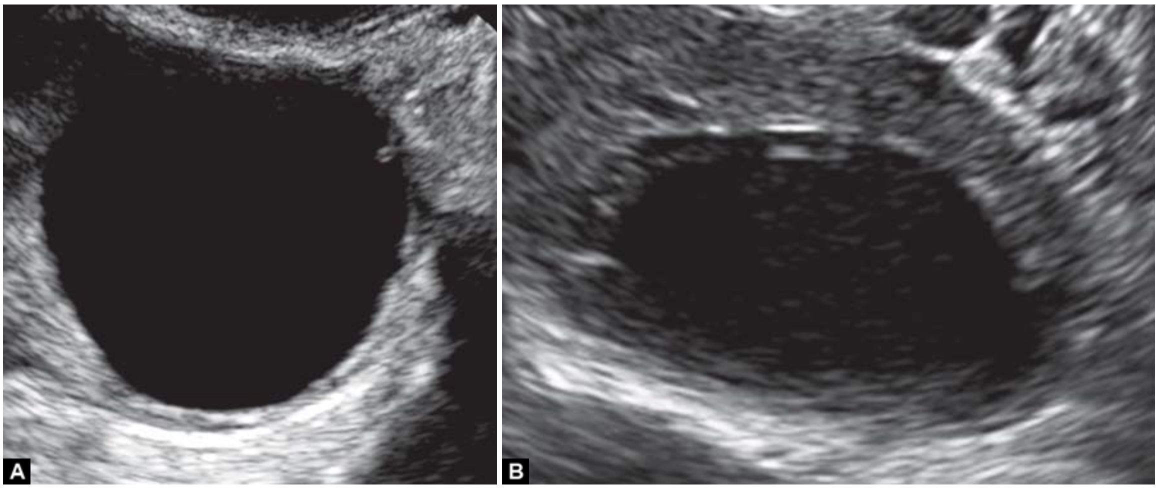
Antral follicles in the early follicular stage of the menstrual cycle start growing under the effect of endogenous or exogenous FSH. The dominant follicle is selected very early during the menstrual cycle and becomes apparent as early as 5th day on US. Many a times the follicle that appears to be a leading follicle on day 5 scan may not be the follicle that would ultimately grow and give ovum. The fate of the follicle can be decided by size, rate of growth (as high as >2.3 mm/day in conception cycles) internal echogenecity and wall thickness.

A follicle that is of >10 mm in diameter grows at a rate of 2 to 3 mm per day, has no internal echogenecity and has thin (pencil line like) walls is not only more likely to become the leading follicle but will also give mature healthy ovum.

The growing follicle can be assessed by transvaginal sonography. The shape changes from round to polygonal when there are multiple follicles due to pressure effect from adjacent follicles. In such follicles, the follicular volume would be considered instead of a single diameter and is calculated as (D1 x D2 x D3) × 0.523, where D1, D2 and D3 are three diameters of follicles in perpendicular planes using B-mode US. The follicle shape may become ellipsoid, if pressure is applied by the transvaginal probe on the follicle and, therefore, scans should be done with only optimum pressure applied by the probe.

B-Mode Features of a Mature Follicle

The follicular diameter is measured when the follicle is seen as a rounded structure, or at least three measurements must



Figs 1A and B: (A) Preovulatory follicle on B-mode, (B) 6 to 8 years before rupture

be taken perpendicular to each other and take a mean measurement as follicular diameter. A mature follicle is 16 to 18 mm (Fig. 1A), has thin walls, regular round shape and no echogenecity in the lumen and shows a thin hypoechoic rim surrounding the follicle and sometimes (about 35%-40%) cumulus-like shadow may be seen. This thin hypoechoic rim and cumulus-like shadow develops approximately 36 hours before rupture. A flimsy irregular line is seen inside the follicle parallel to the wall about 6 to 8 hours before rupture (Fig. 1B).

Doppler Features of a Good pre-hCG Follicle

Increase in perifolliclular vascularity of dominant follicle in theca layer starts developing as early as 8th day of the cycle. Fall in perifollicular RI starts 2 days before ovulation, reaches nadir at ovulation, remains low for 4 days and then with gradual rise reaches 0.5 in midluteal phase.[2] When functionally mature, on color Doppler, the follicle shows blood vessels covering at least 3/4th of the follicular circumference (Fig. 2). Chui et al graded the follicular flow on the day of oocyte collection as grade 1 to 4 when in a single cross area slice the flow covered <25, 25 to 50, 50 to 75 > 75% of follicular circumference. The conception was related to grade 3 to 4 vascularity.[3]

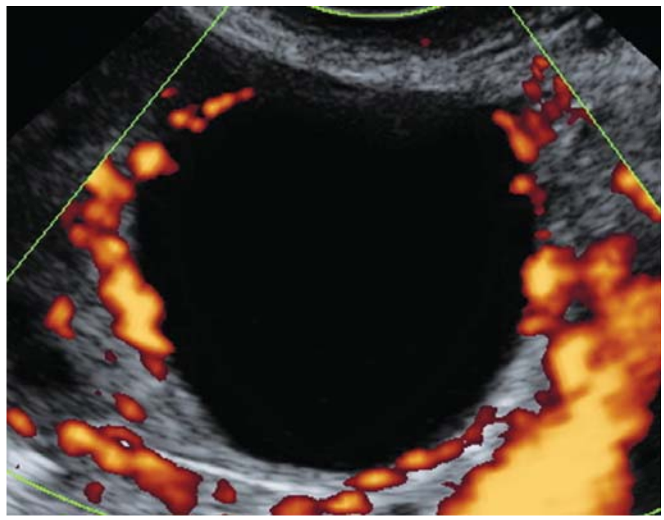


Fig. 2: Preovulatory follicle with vascular ring surrounding the follicle

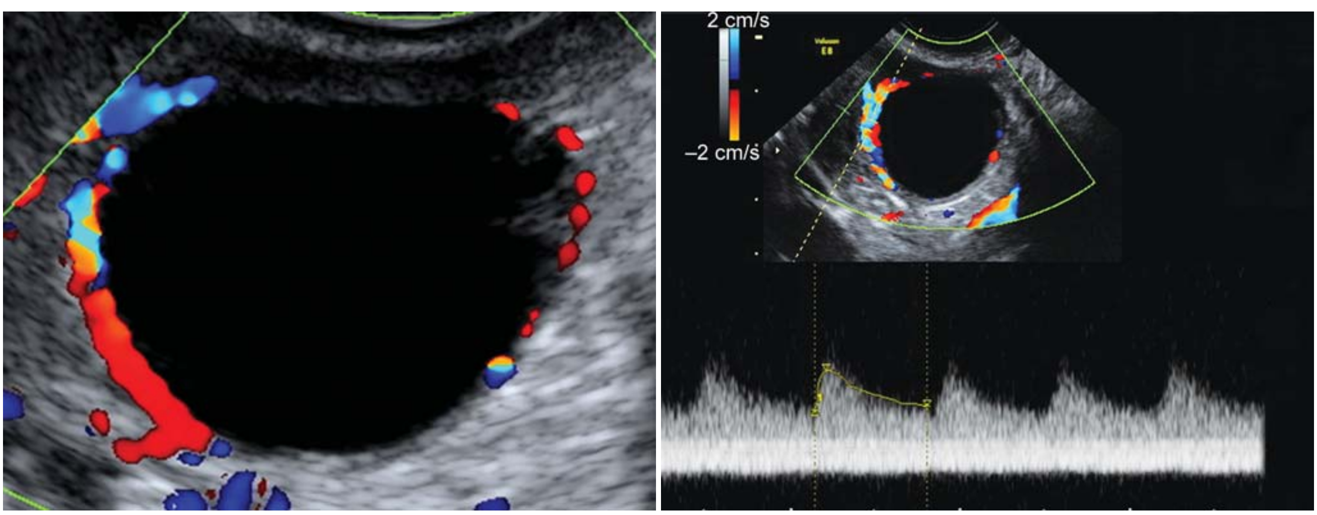


Fig. 3: Preovulatory follicle with pulse Doppler showing low resistance flow

On pulse Doppler, these blood vessels show an RI of 0.4 to 0.48[4] and PSV of >10 cm/sec (Fig. 3). The PRF settings for color Doppler are set at 0.3 and wall filter at the lowest. The perifollicular vessels are only those that obliterate the follicular wall with color. If the follicular wall is seen and the vessel is seen just beside it, it is not a perifollicular vessel.

Implications of Flow Parameters on Ovum Quality

Ovarian flow correlates well with oocyte recovery rates and hence may be useful in determining the most appropriate time to administer hCG to optimize recovery rate. Higher RI indicates higher resistance flow to the follicle meaning lower flow during diastolic phase and so reduced phasic oxygen supply to the ovum and hence lack of maturity. Lower PSV again indicates lower blood supply and hence lack of maturity. It has been quoted in a study by Nargund et al[5,6] that embryos produced by fertilization of the ova obtained from the follicles, which had a perifollicular PSV of <10 cm/sec, are less likely to be grade I embryos and also have higher chance of chromosomal malformations. In the same study, it has been shown that the probability of developing a grade 1 or 2 embryo is 75%, if PSV was >10 cm/sec, 40% if PSV was <10 cm/sec, 24% if there was no perifollicular flow. There is yet another study that supports this finding. Oocytes from severely hypoxic follicles are associated with high frequency of abnormalities of organization of chromosomes on metaphase spindle and may lead to segregation disorders and catastrophic mosaics in embryo.[7]

Our Data

Our unpublished data of more than 1000 IUI cycles has shown that when the perifollicular RI > 0.53 and PSV < 9 cm/sec, 12 hours before hCG injection, the conception rates were only 8.3% and 10% respectively as compared with 32.8% and 28.2% respectively and individually when perifollicular RI <0.50 and PSV >11 cm/sec.

Follicular RI: (1025 cases)

Foll. RI	0.56-0.53	0.53-0.50	<0.50
Conc.	05	71	170
NonConc.	58	373	348
Total	63	444	518

Conc.: Conceived cases; NonConc.: Nonconceived cases

Follicular PSV: (1025 cases)

Foll. PSV	<9	9-10	10-11	> 11
Conc.	04	14	38	190
NonConc.	36	92	168	483
Total	40	106	206	673

Conc.: Conceived cases; NonConc.: Nonconceived cases

We have, therefore, always preferred to wait with no extra medication when patient is on clomiphene citrate stimulation or continue with the same dose of gonadotrophin till we get proper perifollicular RI and PSV, though sometimes the follicular size may reach up to 22 mm. Almost 90% of the times the desired RI and PSV are reached by the time follicular size is 20 mm maximum.

Volume Ultrasound for Follicular Assessment

We use 3D USG and 3D power Doppler for the assessment of these follicles. We studied 500 IUI cycles that were all monitored for ovulation by 2D and color Doppler. When the follicles appeared mature as per the above-mentioned features, 3D and 3D power Doppler was done for all and the values of 3D and 3D power Doppler indices were studied and plotted on the graphs for conception and nonconception cycles. We used Voluson 730 Expert (GE) for our studies. Once the follicles of ≥6 mm for gonadotropin stimulation or ≥18 mm for cc stimulated cycle is seen and assessed by color Doppler, the volume box is switched on. The size of the volume box should be enough to include the whole follicle and at least a surrounding 5 to 7 mm margin. The angle of the volume should be large enough to cover the whole follicle from edge to edge. The acquired volume is seen as follicle in three perpendicular planes on the screen. Using the vocal software with 15° angle the follicle is traced

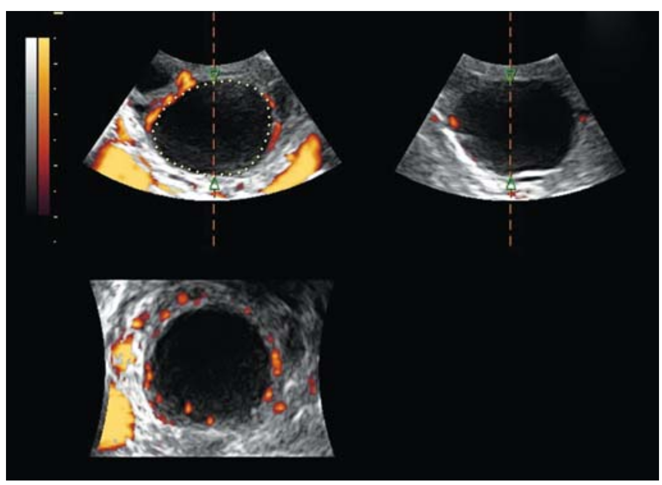


Fig. 4: Calculating follicular volume by vocal

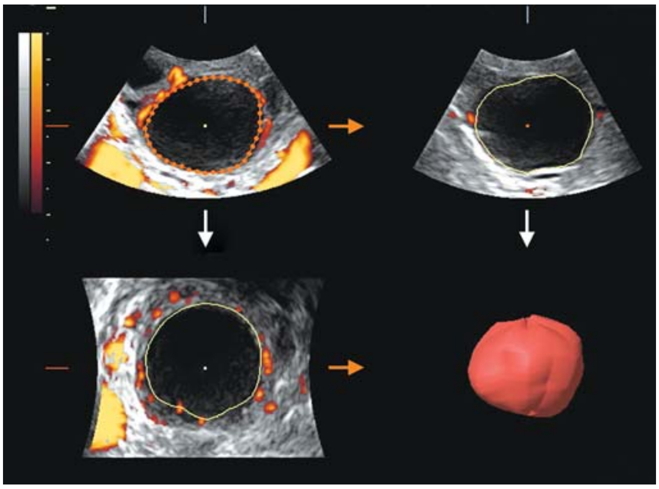


Fig. 5: Follicular volume by vocal

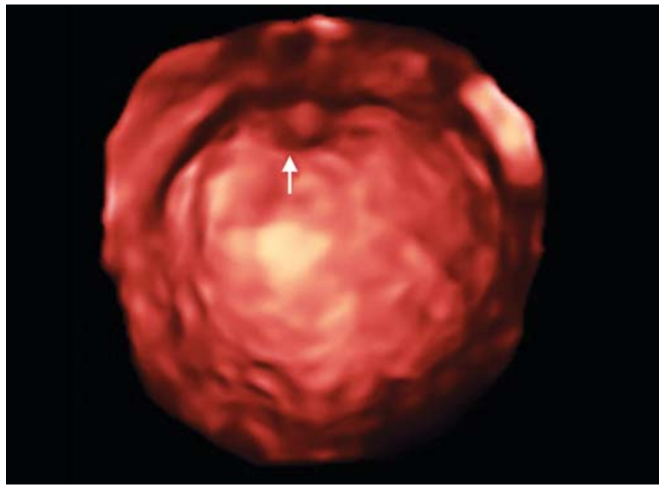


Fig. 6: Cumulus Oophorus in preovulatory follicle

at its circumference at every 15° rotation (Fig. 4) and at the end the command 'done' is given, the volume is ultimately accepted, or any corrections required may be made and the machine then calculates the volume of the follicle.

Volume Ultrasound Parameters of Good Pre-hCG Follicle

Follicular Volume

On 3D the follicular volume of 3 to 7.5 cc has been found to be optimum in our study[34] (Fig. 5). This agrees with the study by Witmack et al[8] which says that : In IVF -ET cycles, follicles with mean follicular diameter of 12 to 24 mm are associated with optimal rates of oocyte recovery, fertilization and cleavage. This corresponds to the follicular volumes of between 3 and 7 ml. The accuracy of 3D USG measurement of follicular volume compared to the standard 2D techniques by comparing the volume of individual follicles estimated by both methods with the corresponding follicular aspirates: Using the formula of ellipse the limits of agreement between aspirates and calculated volume were +3.47 to –2.42 as compared with +0.96 to –0.43 when calculated by 3D US using VOCAL.[9] This is because 3D US measurement is not affected by the follicular shape as the changing contours are outlined serially to obtain the specific volume measurement. Though the follicles of <10 mm in diameter, cannot be assessed accurately by 3D US because limits of agreement are too wide in this range.

Cumulus

After the volume calculation the follicle is seen plane by plane in the acquired volume by translation and rotation, that is walking through the volume and rotating the volume. Or one can also use tomographic ultrasound imaging (TUI), in all three planes, with a slice thickness of 0.5 mm, zoom the image so that only the follicle can be seen on each image and slices are examined one by one or in a set of 4 for the presence of cumulus like echo. If it is seen on one plane it is confirmed on the other two planes and also on the rendered image. Using a combination of surface smooth and light gradient mode for rendering with high threshold can show beautiful cumulus (Fig. 6). We in our study of 500 IUI cycles have been able to locate the cumulus in 94.6% of cases in conception cycles and in 53% of nonconception cycles by surface rendering usually and TUI (0.5 mm slices) sometimes. In IVF cycles also, we have been able to predict the number of ova that we would obtain on retrieval by the number of cumulus containing follicles. Feichtinger et al in their study have shown presence of cumulus in follicles >15 mm by 3D US.[10] Follicles without visualization of cumulus in all three planes are not likely to contain mature oocytes. Poehl et al also showed in their study that appearance of the intrafollicular cumulus structures by 3D US was correlated with the recovery rate of the mature oocytes.[11] They also found a significant correlation between the number of detected cumuli and the number of retrieved oocytes ($p < 0.0001$), mature oocytes ($p < 0.0001$) and number of fertilized oocytes ($p < 0.0001$). Therefore, visualization of the cumulus by 3D US is a positive indicator of mature oocytes in both IUI and IVF procedures and has been found to be an indicator of successful fertilization in IVF cycles also.

3D Pulse Doppler for Follicular Assessment

It has been shown that follicular fluid concentrations of leptine, a follicular angiogenesis-related factor, are inversely related to the stromal blood flow index.[12] It has also been suggested that the follicles containing oocytes capable to produce a pregnancy have a perifollicular vascular network more uniform and distinctive.[13]

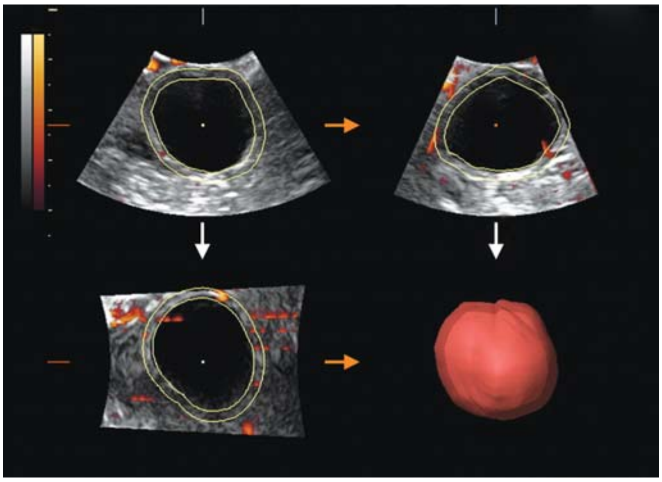


Fig. 7: Calculating the shell volume of the follicle for perifollicular 3D power Doppler assessment

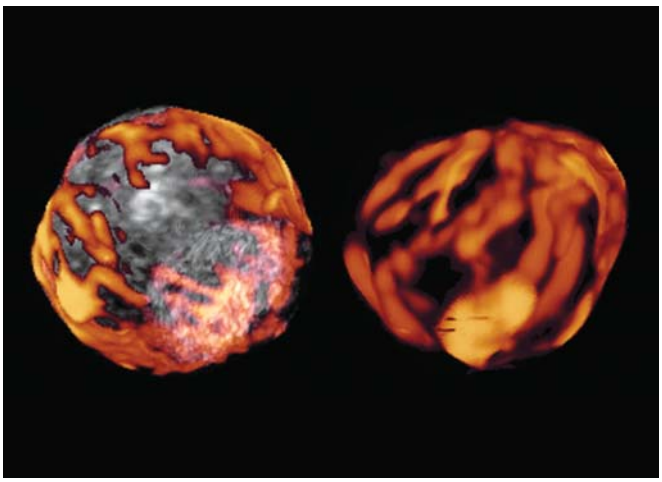


Fig. 8: Angiomode of the perifollicular vascularity

Fig. 9: Histogram for perifollicular flow giving VI, FI and VFI values

After having measured the volume of the follicle and looking for the presence of the cumulus, we switch over to power Doppler. The PRF settings are fixed at 0.3 and wall filter at the lowest. A 3D volume with power Doppler is taken, the volume angle same as that for the follicular volume is selected. Again on the screen, the follicle is seen in three perpendicular planes but this time with blood vessels as seen on power Doppler. Again switch on the vocal and calculate the follicular volume. At the end of the last scroll around the follicle, click the 'done' switch and the options are open to accept the ROI or to select one of the options for the shell volume. We select an option for the outside shell with the wall thickness of 2 mm, which has been found to be the most appropriate to include the perifollicular vessels (Figs 7 and 8). Then accept this and switch on the volume histogram. When this volume is accepted it gives you the follicular volume as the reference volume, the shell volume as the volume of the wall and the whole combined volume. So for all practical purposes, we usually omit taking the volume of the follicle without the power Doppler first as this vocal calculation with power Doppler will give us the follicular volume also. Moreover, the same volume can be assessed for the cumulus also as described earlier and that saves time spent on each patient for acquiring and calculating one extravolume dataset. So then when we switch over to volume histogram, we get a graph which shows the calculation of the gray voxels and the color voxels in the given volume and these values are displayed on the screen as VI, FI and VFI values. VI indicates the abundance of the color voxels in the given volume, FI indicates the intensity of the color in the given volume and VFI is the ratio of the abundance and intensity, meaning it gives idea about the general perfusion status of the given volume (Fig. 8). This means volume histogram or the 3D power Doppler indices gives the quantitative assessment of global vascularization and perfusion of the given volume. So after calculating the volume of the follicle and the shell volume when we switch on the volume histogram, the VI, FI and VFI values that we get are the perifollicular vascualrization and perfusion indices. This gives idea about global follicular vascularity rather than interrogation of only one or two vessels, when we use 2D with Doppler.

3D Power Doppler Indices and Follicular Quality

In our study[34] we have found perifollicular VI of between 6 and 20 and perifollicular FI > 35 as most optimum. A total of 68.4% of patients conceived when the VI was between 6 and 18 and 50% when it was between 18 and 20. However, the pregnancy rates were < 25% when VI was <6 and only 7.4% when VI was > 20. It was only 7.4% of patients with FI < 27 who conceived where as beyond 27, the conception rates rose consistently. It was 50% with FI between 27 and 35, 70% when FI was between 35 and 43 and almost all patients had conceived when FI was >43 (Fig. 10).

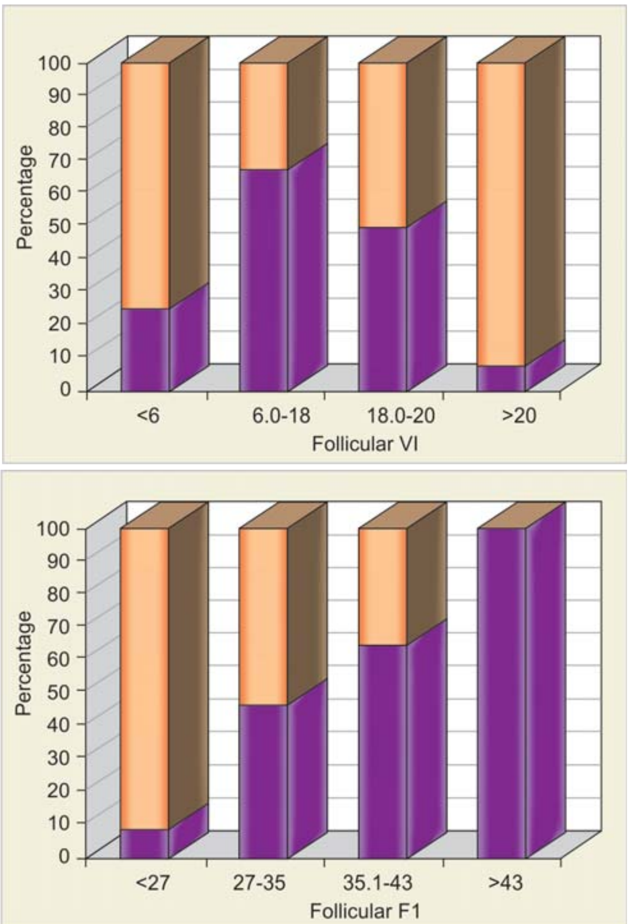


Fig. 10: Pre-hCG perifollicular 3D power Doppler values

Follicular volume: (500 cases)

Foll. vol.	<3	3-5	5.1-7	>7
Conc.	15	94	85	20
NonConc.	46	89	66	95
Total	61	173	151	115

Conc: Conceived cases; NonConc: Nonconceived cases

Follicular VI: (500 cases)

Foll. VI	<6	6-18	18.1-20	>20
Conc.	6	115	83	10
NonConc	20	55	83	128
Total	26	170	166	138

Conc: Conceived cases; NonConc: Nonconceived cases

Follicular FI: (500 cases)

Foll. FI	<27	27-35	35.1-43	>43
Conc.	12	100	76	26
NonConc.	153	100	32	1
Total	165	200	108	27

Conc: Conceived cases; NonConc: Nonconceived cases

Meaning that even when the follicle appeared mature according to the 2D US and color and pulse Doppler parameters, the pregnancy rates were significantly better only when the follicular volume was between 3 and 7.5 cc, cumulus was present and the perifollicular VI and FI values were as mention above.

A study by Kupesic and Kurjak shows that when the ratio of follicular volume to blood flow index (FV/FI) is between 0.4 and 0.6 the pregnancy rates are 39%, if >0.6, it is 52% and when <0.4 is only 21%.[14]

This explains us the failure to achieve pregnancy in IUI cycles and failure to retrieve ova or achieve fertilization in IVF cycles even when hCG is given after proper assessment of follicle by 2D and color Doppler US. Although it is possible to assess the follicular flow as expressed by the peak systolic velocity and perifollicular color map,[15] it is the 3D power Doppler which proves the most precise information about the vascularization and follicular blood flow.[16]

Doppler Parameters of Follicle and Time of Intrauterine Insemination

Going a little further from this stage, hCG plays a major role in inducing influx of blood within follicles. At the LH surge the perifollicular PSV is 10 cm/sec. The follicular PSV goes as high as 45 cm/sec before an hour of ovulation under the effect of rising LH. This means that, if the follicle is said to be functionally mature when PSV is 10 cm/sec, that is the time when the LH surge starts and under the effect of that LH, the perifollicular PSV keeps on rising constantly. This derives that a rising PSV with steady low RI suggests that the follicle is close to rupture.

Based on these facts, we did a study to find out, if double IUI can increase the pregnancy rate in patients who have a pre-hCG perifollicular PSV > 15 cm/sec. A total of 300 IUI cycles were studied, with three different stimulation protocols. The protocols were only clomiphene citrate 100 mg from day 5 to day 9, aromatase inhibitor tablet letrozole 100 mg from day 3 to 7, followed by rFSH (recombinant FSH) 75 iu till the follicle size of 16 to 18 mm was achieved, and only rFSH from 3 of the cycle till the desired follicular size was achieved. All the patients had unexplained infertility, were between 23 and 29 years of age and had a body mass index (BMI) of between 23 and 27. Color and pulse Doppler was done for all patients. Stimulation was continued till optimum flows were also seen apart from the size of the follicle. hCG 10,000 was given to all patients for follicular rupture. Every alternate patient was taken for double IUI, irrespective of color Doppler findings. Single IUI was done between 36 and 38 hours and double IUI were done at 12 to 14 and 36 to 38 hours postwash sperm count between 7 and 10 million/ml.

The results clearly showed that no matter what protocol was used, the conception rates were significantly high in patients in whom double IUI was done when perifollicular PSV was ≥20 cm/sec on the day of hCG, as compared to when single IUI was done. When perifollicular PSV on the day of hCG was between 15 and 20 cm/sec, the conception rates were only minimally increased with double IUI. Though the conception rates were similar for single or double IUI when perifollicular PSV on the day of IUI was <15 cm/sec (Fig. 10).

Endometrial Evaluation

Implantation has been weakest link in the success of infertility treatment. Endometrium is a receptor organ for majority of the hormones involved in fertility and therefore study of its morphology and vascularity is thought to explain the mysteries of implantation failure. Therefore, like follicle, endometrium is also assessed by transvaginal 2D US and color Doppler before planning for hCG during any assisted reproductive technologies.

B-Mode Features of Endometrium with Good Receptivity

On TVS an endometrial thickness of minimum 6 mm is required on the day of hCG, but 8 to 10 mm is optimum. Endometrial thickness has more negative predictive value for implantation. Morphology of the endometrium is as important as thickness of the endometrium. In all the healthy endometria, the endometriomyometrial interface is always seen as a clear hypoechoic halo surrounding the whole endometrium (Fig. 11). Breach or irregularity of endometrio-myometrial junction is an indication of unhealthy endometrium and therefore poor receptivity.

Popularly multilayered endometrium is considered as a desired endometrial pattern. Morphologically, the endometrium is graded as the best grade A, when it is a triple line endometrium with the intervening area is as hypoechoic as the anterior myometrium. The echogenecity is attributed to the development of multiple vessels penetrating in the endometrium producing multiple tissue

Fig. 12: Most receptive endometrium grade A—multilayered

Fig. 13: Multilayered endometrium grade B—intermediate

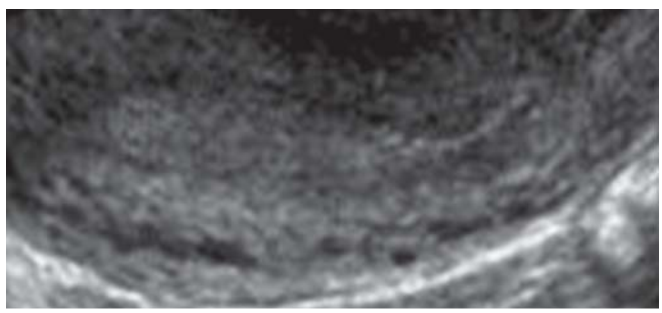


Fig. 14: Isoechoic homogenous grade C—endometrium

Fig. 11: Endometriomyometrial junction

interfaces and therefore causing the echogenecity and due to glycogen storage in the endometrial columnar epithelium (Fig. 12). The endometrium is graded as intermediate or grade B (Fig. 13) when it is multilayered or triple line with hypoechoic intervening area. In grade C or the most unfavorable endometrium would be a homogenous isoechoic endometrium[17] (Fig. 14). Though some studies have shown no significant difference in pregnancy rates among different morphological patterns.

Doppler Features of Endometrium with Good Receptivity

There are several reports by different groups[18] that agree on the fact that implantation rates can be more correlated to the vascularity of the endometrium rather than the thickness and morphology of the endometrium.

Segmental uterine artery perfusion demonstrates significant correlation with hormonal and histological markers of uterine receptivity, reaching the highest sensitivity for subendometrial blood flow.[14] On color Doppler, the endometrium which is mature shows vascularity in zone 3 and 4 or may be called subendometrial and endometrial layers (Fig. 15). The zones of vascularity are defined according to Applebaum[19] as: Zone 1 when the vascularity on power Doppler is seen only at endometrio-myometrium junction, zone 2 when vessels penetrate through the hyperechogenic endometrial edge, zone 3 when it reaches intervening hypoechogenic zone and zone 4 when they reach the endometrial cavity. The pregnancy rates related to the zones of vascular penetration: 26.7% for zone 1, 36.4% for zone 2 and 37.9% for zone 3.

One more comparision of two studies have also shown similar results[20]:

- Zone 1 3.5-7.5% 5.2%
- Zone 2 15.8-29.7% 28.7%
- Zone 3 24.2-47.8% 52%
- Zone 4 67.3 74%

Though in our unpublished data of more than 1000 IUI cycles, when color Doppler studies were done 12 hours before hCG injection, we have found only 7.3% pregnancy rates for zone 1 and 13.4 % for zone 2 vascularity. The conception rates with zone 3 and 4 the pregnancy rates were comparable and were 35.8% and 38.3% respectively.

Zone of vascularity: (1025 cases)

Vascular zone	1	2	3	4
Conc.	10	52	100	84
NonConc.	125	336	181	137
Total	135	388	281	221

Conc.: Conceived cases; NonConc.: Nonconceived cases

Zaidi et al found that absence of flow in the endometrial and subendometrial zones on day of hCG indicate total failure of implantation.[18] The vessels that reach the endometrium are the spiral arteries. The pulse Doppler of these arteries should have an RI of between 0.6 and 0.8 and PI of between 1.1 and 2.3 for the endometrium to be called mature for implantation (Fig. 16).

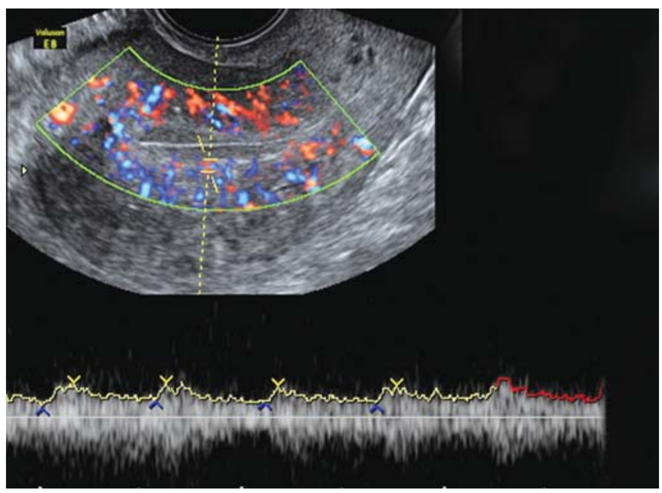


Fig. 16: Pulse Doppler of endometrial vascularity

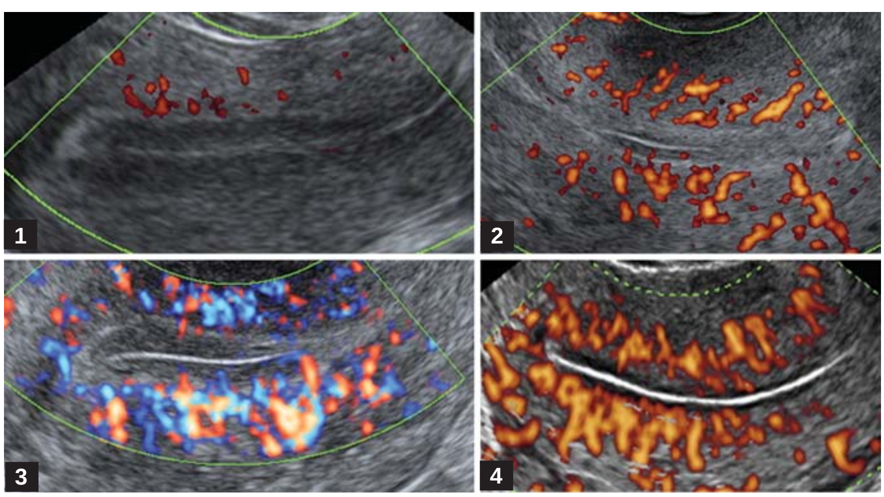


Fig. 15: Endometrial vascularity zones

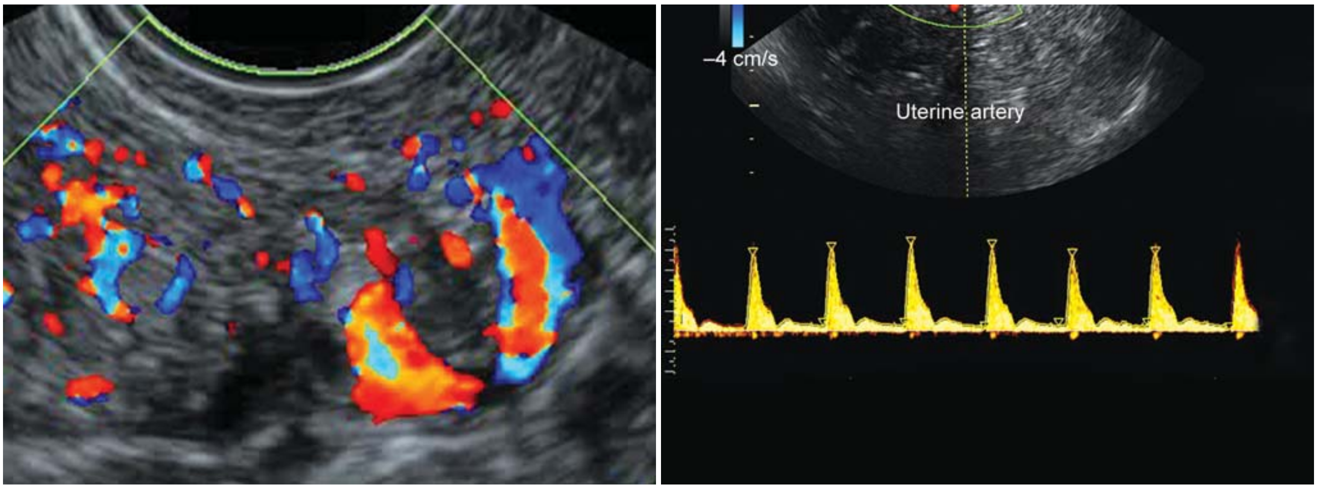


Fig. 17: Preovulatory uterine artery waveform

Uterine Artery Doppler

Moreover, the pulse Doppler analysis of the uterine artery waveform is done and its RI should not be more than 0.9 and PI should be <3.2 (Fig. 17). Several authors have shown that the optimum uterine receptivity was obtained when average pulsatility index of the uterine artery was between 2 and 3 on the day of transfer or on the day of hCG.[21,22] Coulam et al have also shown that no pregnancy was achieved after embryo transfer when uterine artery PI was above 3.3 in an IVF program.[23] Tsai et al[24] evaluated the prognostic value of uterine perfusion on day of hCG for IUI cycles and showed that no pregnancy occurred when the pulsatility index of ascending branch of uterine artery was >3. Fecundity rate was 18% when PI < 2 and 19.8% when between 2 and 3. This probably can be explained on the basis that the uteri having fibroids or adenomyosis show fairly high uterine artery blood flow velocities but lower resistance indices. A study by Cacciatore et al suggest that implantation is unlikely when PI is >3.3 and RI > 0.95 or when no velocities are seen at the end of the diastole.[25] Though uterine artery impedance is usually low in superovulated cycles. In our unpublished data of more than 1000 IUI cycles, when color Doppler studies were done 12 hours before hCG injection, we have found no conceptions when uterine artery PI > 3.5.

Volume Ultrasound Assessment of Endometrium

Then when endometrial and the uterine artery parameters are optimum as described above, the volume of the endometrium with power Doppler is acquired and then the endometrial volume is calculated by vocal, tracing the outer edge of the hyperechoic outer rim of the endometrium and accept the volume (Fig. 18). The endometrium up to the internal os is taken into calculation. For accurate measurements, a good contrast in the image is necessary. Select smaller angles of rotation for VOCAL. Fluid in the endometrial cavity can be a source of error. For endometrial volume the inter-CC definition of internal os and (interobserver variation) was 0.82 and intra-CC (intraobserver variation) was 0.90, the chief source of error being definition of endometrial margins.[26]

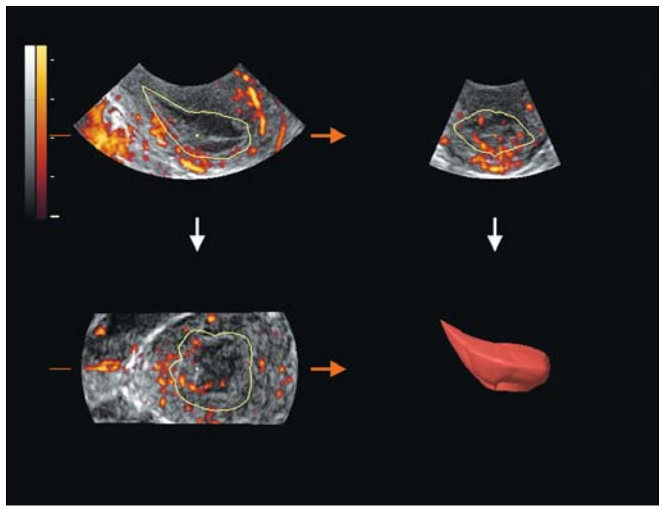


Fig. 18: Calculating endometrial volume by vocal

3D and 3D Power Doppler Features of Good Endometrial Receptivity

Our study[34] showed that at endometrial volume of <2 cc no pregnancies occurred. With endometrial volume of 2 to 3 cc only 16.66% of patients conceived, between 3 and 5 cc 47% and when the endometrial volume was between 5 and 7 cc 61.5% patients conceived.

Endometrial volume: (500 cases)

Endo. vol	<2	2-3	3.1-5	5.1-7	>7
Conc.	0	26	70	67	41
NonConc.	20	132	78	42	24
Total	20	158	148	109	65

Conc.: Conceived cases; NonConc.: Nonconceived cases

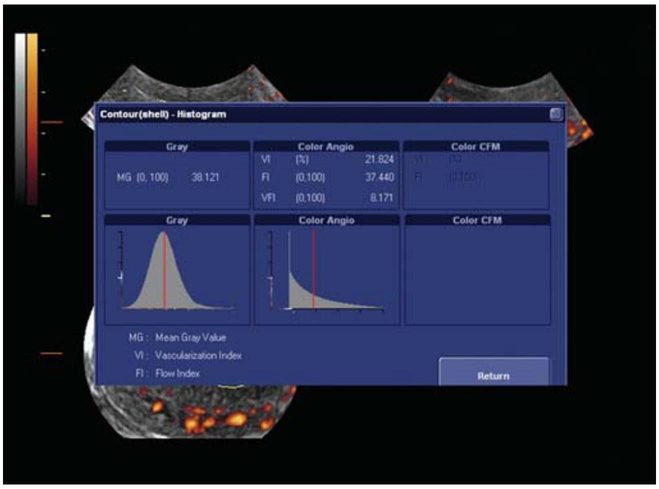


Fig. 19: Histogram of endometrium showing VI, FI and VFI values

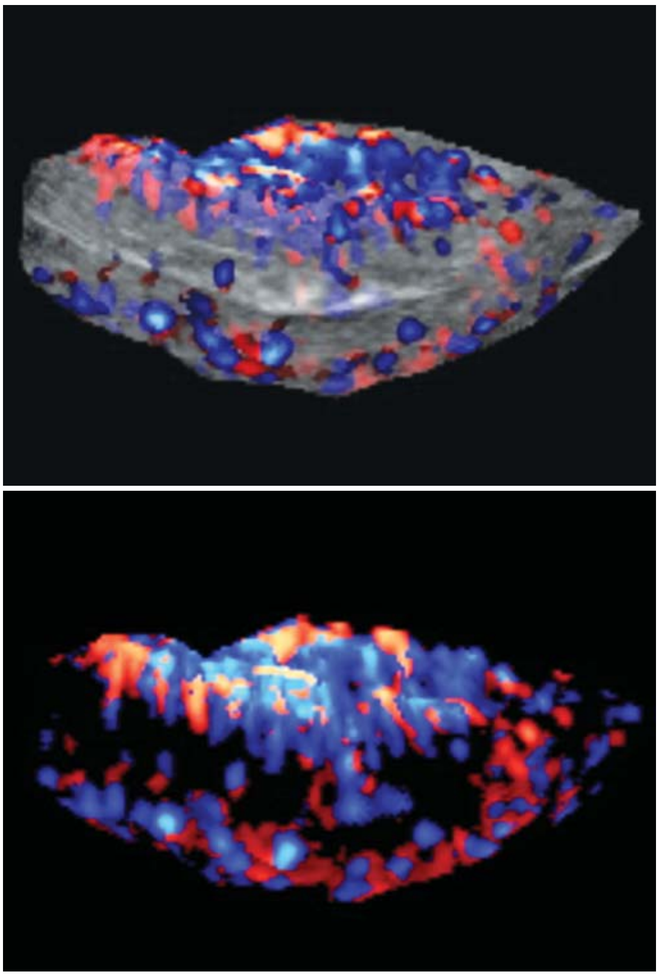


Fig. 20: Glass body mode of endometrial 3D power Doppler vascularity

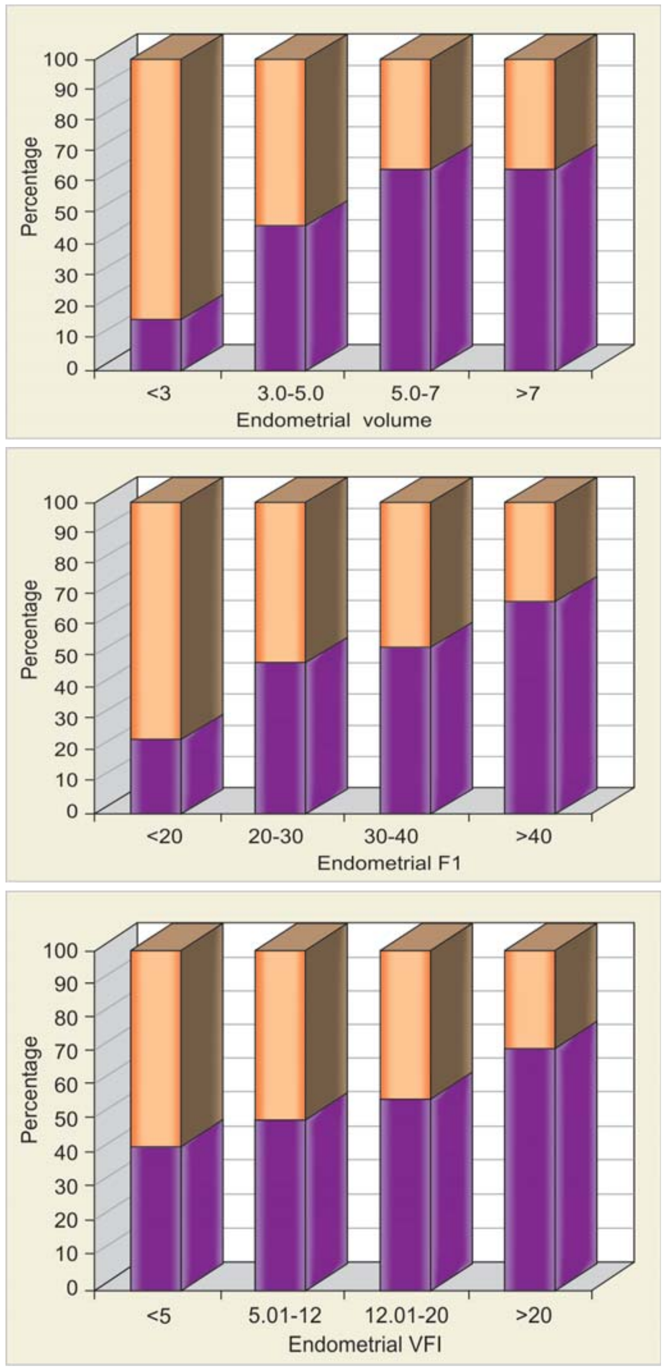


Fig. 21: Pre-hCG endometrial volume and 3DPD values

Decline in conception rates at endometrial volumes of more than 7 cc is seen. This can be explained by pathological increase in endometrial thickness and volume like hyperplasia or large polyps, that significantly decrease the implantation rates. Endometrial volume by 3D US volume calculation of the endometrium may help to correlate the cycle outcome with quantitative parameter rather than endometrial thickness.[8] A study by Raga et al[27] shows pregnancy and implantation rates were significantly lower when endometrial volume <2 ml while no pregnancy was achieved when endometrial volume was <1 ml. Study by Kupesic et al also shows no pregnancy when endometrial volume was <2 ml or when exceeded 8 ml.[28] For the same calculated volume, volume histogram is switched on and it calculates the endometrial VI, FI and VFI (Figs 19 and 20). In our series[34], the endometrial VI values were not very conclusive. Endometrial FI when was <20, only 23% of patients showed conception, between 20 and 40, it was almost 50% but when FI was more than 40, 68% of patients showed conception.

Endometrial FI: (500 cases)

Endo. FI	<20	20-30	30.1-40	>40
Conc.	37	68	75	34
NonConc.	125	76	68	17
Total 162	144	143	51	

Conc.: Conceived cases; NonConc.: Nonconceived cases

Endometrial VFI when was >20 the conception occurred in 71.2% of patients and when VFI was <1.0 no conception was seen. Though between VFI 5 and 20 the percentage of conception was 49% to 56% (Fig. 21).

Endometrial VFI: (500 cases)

Endo.VFI	<2	2-5	5.1-12	12.1-20	>20
Conc.	00	96	64	45	09
NonConc.	44	136	66	36	04
Total	44	232	130	81	13

Conc.: Conceived cases; NonConc.: Nonconceived cases

A scoring system reported by Kupesic et al[28] for uterine receptivity, done on the day of embryo transfer shows that subendometrial FI < 11 was a cut off limit (Subendometrial indices are calculated in the same way from the endometrial volume as we calculate the perifollicular indices from the follicular volume, i.e. by shell volume of 2 mm thickness). No pregnancies occurred when it was <11 and the conception group showed its values of 13.2 $\pm$ 2.2. These values differ from our values probably because we have done calculations on whole endometrium as compared with the calculations made for the subendometrial layers here, but is common in both that higher FI values are associated with higher pregnancy rates.

Whereas Ng et al[29] documented a low endometrial VI and VFI in pregnant group on the day of oocyte retrieval and also a nonsignificant trend of higher implantation and pregnancy rates in patients with absent subendometrial and endometrial flow. This probably can be explained on the basis that hCG administration/LH peak causes increased uterine artery resistance and hence decrease in endometrial perfusion also on the day of oocyte retrieval. This also correlates with the observation made by Ng et al[30] which says that subendometrial vascularization flow indices are significantly lower in patients with uterine artery RI $\geq$ 0.95. They concluded that number of embryos replaced and endometrial VFI was the only two predictive factors for pregnancy. Wu et al[31] reported that endometrial VFI was more reliable than VI and FI, and best prediction rate was achieved by VFI cut off value of >0.24.

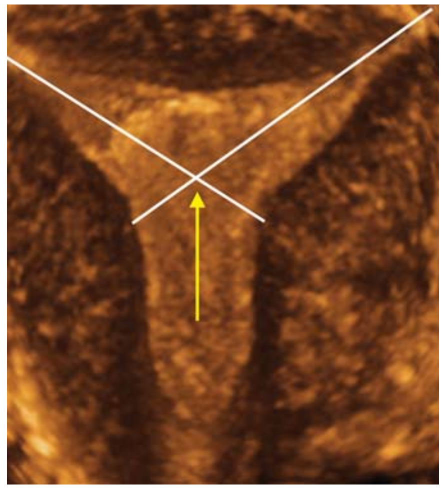


Fig. 22: Maximum implantation point

Contractions and MIP

Apart from these criteria, there are another few that have been worked upon in some studies. It has been shown that uterine motility on the day of embryo transfer has an impact on implantation rates. Number of uterine peristaltic contractions is counted per minute in longitudinal scan of the uterus. More than 5 contractions per minute has considered as unfavorable sign and <3 has been considered as a favorable sign for implantation.[32] Point of maximum implantation also has been worked out. This point is an intersection of two lines that are drawn parallel to both cornu on a mid true coronal section of uterus[33] (Fig. 22).

SUMMARY

	Follicle	*Endometrium*
Size/thickness	16-18 mm	8-10 mm
Morphology	Thin wall, no internal echoes, halo	Grade A/B
Vascularity	3/4th circumference	Zone 3-4
RI	0.4-0.48	<0.5
PSV	>10 cm/sec	—
Uterine artery PI	—	<3.2
volume	3-7 cc	3-7 cc
3D morphology	Cumulus	Intact endo-myo junction
3D power Doppler	More symmetrical the better	Higher the better

3D ultrasound is much accurate for volume assessment both for follicle and the endometrium, and are much more reliable parameters than follicular diameter or endometrial thickness. The presence of cumulus can be confirmed by 3D US increases the surety of the presence of a mature ovum in the follicle. The 3D power Doppler gives idea about the global vascularity of follicle and endometrium. Though still larger studies and standardization of ultrasound parameters and settings are required to establish more precise values for follicular and endometrial VI, FI and VFI, the results are fairly promising. We can hope to understand the follicular and endometrial physiological status better with these parameters and achieve better pregnancy rates with ART procedures and reduce the span of unexplained infertility.

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