Three-dimensional ultrasound features of the polycystic ovary and the effect of different phenotypic expressions on these parameters

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BACKGROUND: The aim of this study was to quantify the three-dimensional (3D) ultrasound characteristics of ovaries in Caucasian women with polycystic ovarian syndrome (PCOS) and to examine if these values differed between different phenotypic forms. METHODS: 3D pelvic ultrasound was performed in 40 women with PCOS and in 40 controls. Total ovarian volume, stromal volume and echogenicity and antral follicle count (AFC) were measured and ovarian blood flow was quantified using both 3D power Doppler and two-dimensional pulsed-wave Doppler. RESULTS: Women with PCOS had a higher AFC (median 16.3 versus 5.5 per ovary, \(P<0.001\)) and ovarian volume (12.56 versus 5.66 ml, \(P<0.001\)). Stromal volume (10.79 versus 4.69 ml, \(P<0.001\)) and stromal vascularization (VI: 3.85 versus 2.79\%, \(P<0.001\); VFI: 1.27 versus 0.85, \(P<0.001\)) were also increased in women with PCOS. There were no significant differences in stromal echogenicity or pulsed-wave Doppler indices between women with PCOS and the controls. Among the women with PCOS, ovarian vascularity was significantly higher in 30 women who were hirsute compared with normoandrogenic women (FI: 33.94 versus 29.30, \(P<0.05\)) and in 14 women with PCOS who were of normal weight compared with obese women (VI: 4.51 versus 3.25\%, \(P<0.05\); VFI: 1.56 versus 1.22, \(P<0.05\)). CONCLUSIONS: Based on 3D ultrasound, women with PCOS have an increased stromal volume and vascularity. Stromal vascularity is significantly higher in women with PCOS who are hirsute and of normal weight.

Keywords: polycystic ovary; PCOS; three-dimensional ultrasound; ovarian volume; ovarian vascularity

Introduction

Polycystic ovarian syndrome (PCOS) is the most common reproductive endocrinopathy of women during their childbearing years, with a reported prevalence of 5–10\% (Franks, 1995). As a heterogeneous disorder with variable manifestations and subjective diagnostic criteria, studies of women with PCOS have not been uniform and comparative analyses are open to criticism. This lead to a consensus in 2003 when a joint meeting of the European Society for Human Reproduction (ESHRE) and the American Society of Reproductive Medicine (ASRM) established new guidelines for the diagnosis of PCOS (The Rotterdam ESHRE/ASRM-Sponsored PCOS consensus workshop group, 2004). The new ‘Rotterdam criteria’ allow a diagnosis of PCOS to be made when two of three clinico-pathological features are present: oligomenorrhoea or anovulation, clinical or biochemical hyperandrogenism and clearly defined polycystic ovaries (PCOs) on ultrasound. This revised classification supports the objective role of ultrasound in the diagnosis of PCOS which should include either 12 or more follicles measuring 2–9 mm in diameter or an increased ovarian volume >10 cm³ in either ovary (Balen et al., 2003).

However, there are certain limitations in these ultrasound criteria that should be considered in future work (Lam and Raine-Fenning, 2006). While an objective measurable increase in the number of antral follicles is possible, the real-time interpretation of two-dimensional (2D) ultrasonography may underestimate the absolute number of follicles compared with three-dimensional (3D) ultrasound (Allemand et al., 2006). Furthermore, ovarian volume calculations based on 2D measurements are less accurate and less reproducible than those made with 3D ultrasound (Raine-Fenning et al., 2003a). Several other ultrasound features that were previously considered important, such as an increase in ovarian stromal echogenicity and vascularity, have not been included in the new diagnostic criteria. This probably relates to the inherent subjectivity in the objective or semi-quantitative assessment of these parameters and a resultant inability to standardize measurements (Balen et al., 2003). 3D ultrasound provides a new...
method for the objective quantitative assessment of all of these parameters as well as blood flow within the ovary as a whole (Raine-Fenning et al., 2003b; Raine-Fenning et al., 2004a).

Previous studies on the 3D ultrasound features of PCO have consistently demonstrated the intrinsic characteristics of a polycystic ovary confirming an increased antral follicle population and larger ovarian volume (Pan et al., 2002b). However, conflicting results on the degree of vascularity within a polycystic ovary as a whole or within its stroma have been observed and the study with the most scientifically robust design did not demonstrate the widely held belief that stromal vascularity is increased (Ng et al., 2005). It has been suggested that the variation between studies may relate to different characteristics of the study population, the use of inappropriate controls and inconsistent criteria for the diagnosis of PCOS (Lam and Raine-Fenning, 2006). We have conducted a prospective observational study in women with PCOS, as defined by the Rotterdam criteria, to examine this hypothesis and used 3D ultrasonography to examine the ultrasound features in women with different phenotypical manifestations of PCOS compared with a control population.

Materials and methods
This prospective observational study was approved by National Health Service Research Ethics Committees in UK. Information leaflets outlining the study were given to the potential participants along with the standard information sheet on PCOS. Any woman considering entering the study was referred to one of the investigators (P.M.L.) who described what was involved and obtained written consent.

Subjects
We aimed to recruit 40 Caucasian women with PCOS from the Assisted Conception Unit (NURTURE, Nottingham University Research and Treatment Unit in Reproduction) and the gynaecology clinics at Queen’s Medical Centre as the study group. Inclusion criteria were based on the Rotterdam consensus in that the woman had to have at least two of the following three features: PCOs on ultrasound scan, oligomenorrhoea and/or anovulation and clinical or biochemical hyperandrogenism. Exclusion criteria included any identifiable endocrine cause of anovulation and hyperandrogenism. For the control group, we aimed to recruit 40 subfertile Caucasian women without PCOS, undergoing treatment cycles of intrauterine insemination (IUI) and/or ovulation induction (OI) for infertility due to male factor or unexplained infertility. To be included they had to have regular menstrual cycles (range of cycle length 25–35 days with discrepancy ≤7 days) and biochemical confirmation of ovulation as demonstrated by a mid-luteal serum progesterone level above 28 nmol/l. They also had normal serum levels of follicle stimulating hormone (<10 IU/l) and luteinizing hormone (<10 IU/l) during the early follicular phase of the menstrual cycle. Detailed transvaginal ultrasound scans were performed to exclude pelvic pathology such as hydrosalpinges, adenomyosis and fibroids. Subjects were excluded if they had clinical hirsutism or acne and if there was ultrasonographic evidence of PCOs. Exclusion criteria for both groups included the presence of any ovarian follicles measuring 10 mm or more in diameter or an ovarian cyst on baseline scan, previous ovarian surgery and the use of exogenous hormones in the past 3 months.

The sample size calculation was based on the reported differences in ovarian stromal blood flow between women with PCOS and controls without PCOS [vascularization index (VI): 3.99 ± 2.38 versus 1.44 ± 1.20%; flow index (FI): 50.26 ± 3.02 versus 44.44 ± 5.42; and vascularization flow index (VFI): 2.10 ± 1.32 versus 0.80 ± 0.97; all P < 0.05] (Pan et al., 2002a). In order to detect such differences, a sample size of 18 subjects in each group will give a power of 90% and alpha of <0.05 (Machin et al., 1997). A larger sample size gave a higher power of the study for the same alpha error.

Outcome measures
Clinical parameters
The clinical parameters including the subject’s age, ovulation status, degree of hirsutism and acne and BMI were recorded. Hirsutism was subjectively assessed by patients with a 4-point scale; no clinical hirsutism (0), hirsutism requiring occasional shaving less than once per week (1), hirsutism requiring regular shaving at least once per week (2) and severe hirsutism requiring regular shaving more than once per week and distressing to the patient (3). Acne was also scored similarly by patients; no obvious acne (0), mild acne with <5 spots on the face (1), moderate acne involving mainly the face with five or more spots (2) and distressing acne involving both the face and trunk (3). This assessment method of hirsutism and acne is convenient and easy to use in the clinical setting and provides an indication of the clinical significance of these cosmetic symptoms to the patients. Body weight and height were measured and BMI was calculated from the formula [BMI = body weight in kilograms/(body height in metres)²].

Data acquisition
Ultrasound scans were performed between Days 3 and 5 of the menstrual cycle or after withdrawal bleeding had been induced by a short course of progesterone (10 mg of oral medroxyprogesterone acetate daily for 7 days) following biochemical confirmation that the subject was not pregnant. Using a Voluson 730TM Expert (GE Health-care, Zipf, Austria) and a 7.5-MHZ transvaginal probe, all scans were performed in a locked, private room between 08:00 and 12:00 am by a single investigator (P.M.L.) to remove inter-observer error.

Our ultrasound technique for data acquisition has been described previously (Raine-Fenning et al., 2003a; Raine-Fenning et al., 2003b). Briefly, this included a routine 2D ultrasound assessment of the pelvis and then the acquisition of volumetric data from each ovary in turn. Each ovary was initially identified and large follicles (>10 mm) and cysts were excluded. The volume mode was then entered and the resultant truncated sector defining the area of interest moved and adjusted and the sweep angle set to 90° to ensure that the whole ovary was obtained. A 3D data set was then acquired using the slow-sweep mode. The resultant multiplanar display was examined to ensure that the complete ovary had been captured with particular emphasis on the ovarian cortex in the C plane. The process was then repeated with medium-sweep mode after power Doppler had been applied to outline the vascular pattern within the ovary. Power Doppler settings were kept constant for each acquisition as follows: pulse repetition frequency 1.0, power 4.0, colour gain 38.4, rise 0.2, persistence 0.8, reject 82 and with wall motion filter set to low 2 and the central frequency set to mid. These settings were found to offer the best compromise between small vessel detection and Doppler artefact (Raine-Fenning et al., 2004a; Raine-Fenning et al., 2003b). If there was apparent artefact, such as typical ‘flash’ artefacts seen with bowel movements, the data set was re-acquired until a subjectively satisfactory image was obtained. The resultant four volume data sets (grey-scale and power Doppler data for both the right and left ovary) were then stored to the hard drive of the machine for subsequent transfer to digital versatile disc.
Pulsed-wave Doppler was then used to derive flow velocity waveforms from the main vessels identified within the ovarian stroma. These vessels were clearly distinct from vessels related to the walls of follicles and were demonstrated with the same 2D power Doppler map used for the 3D acquisition. Angle correction was applied whenever possible and waveforms only stored when the angle was $<30^\circ$ from the incident beam. The waveforms were analysed immediately as there is no option to store such data in a dynamic fashion at present.

**Data analysis**

All data were analysed by one observer (P.M.L.). 4D View (version 5.0, GE Kretz), installed on a personal computer, was used for all 3D data analysis and the multiplanar display was used for all measurement techniques. The number of antral follicles, measuring 2–9 mm, in each ovary was counted in both the A plane (longitudinal image) and B plane (transverse image) and the average value was calculated. Virtual Organ Computer-aided AnaLysis (VOCAL™, GE Kretz) was used for volume calculation and quantification of the power Doppler signal. Our technique has been described in detail before (Raine-Fenning et al., 2003b), but to summarize, we employed the rotational method which involved the manual delineation of the ovarian cortex in 12 planes as the data set was rotated through 180° in a consecutive series of 15° rotation steps until a calculated volume was generated. All measurements were conducted in the B plane (transverse image) as the data set was rotated through the A plane (longitudinal image) as this technique has been proved the most reliable in previous studies (Raine-Fenning et al., 2003a).

Following ovarian volume calculation, 3D image rendering was used to demonstrate intra-ovarian morphology. Image rendering allows the user to modify the image display and maximize the view according to the area or subject of interest. In this study, we used the inversion mode (see Fig. 1) to obtain the best display of the antral follicles with minimal artefact (Raine-Fenning and Lam, 2006). The threshold limit determines which tissue is displayed and can be modified until just hypoechoic fluid-filled structures are displayed. The settings are arbitrary and must be determined for each data set. Following determination of the appropriate threshold limit, the number of voxels above and below this limit can be calculated and displayed allowing determination of stromal volume and total follicular volume, respectively (see Fig. 2).

The VOCAL-imaging program was also used to assess ovarian blood flow through the quantification of the power Doppler signal within the defined volume of interest (VOI). Power Doppler data were displayed about their mean as a histogram which allowed the derivation of three vascular indices through various computer algorithms that assess the number and intensity of the colour voxels within the defined volume. The VI represents the proportion of power Doppler information within the total data set relative to both colour and grey information, providing an indication of the degree of vascularity. The FI reflects the mean power Doppler signal intensity, and is thought to reflect volume flow rate, while the VFI, a composite of the other two indices, is suggested as being representative of tissue perfusion. In addition, the mean signal intensity of the grey voxels is calculated automatically and reported as the mean grey (MG) value which provides an objective representation of the mean tissue density and therefore its apparent echogenicity. The exact relationship of these indices to vascularity, blood flow, perfusion and tissue characteristics remains to be determined and the values are best considered as arbitrary indices similar to those generated through waveform analysis. The application of these indices remains valid as they can be acquired and measured reliably and have been shown to differ significantly between populations (Raine-Fenning et al., 2004c; Raine-Fenning et al., 2004b; Raine-Fenning et al., 2004a; Raine-Fenning et al., 2003b).

**Figure 1:** A multiplanar display of a polycystic ovary, showing the rendered inversion mode which highlights hypoechoic structures such as follicles.
The velocity flow waveforms, obtained through the application of pulsed-wave Doppler to one of the main vessels identified within the ovarian stroma, were analysed by conventional methods to obtain measures of peak systolic velocity (PSV), end-diastolic velocity (EDV), resistance index (RI), and pulsatility index (PI). We also present a new index, the Capacitance Index (CI), measured by calculating the area under the curve for the diastolic component of the waveform but considering the duration of the full cardiac cycle. This index theoretically measures the mean resistance in the diastolic compartment only and has not been described previously. All measurements were made by the manual delineation of the waveform over two consecutive cardiac cycles.

Therefore, the ultrasound outcome measures included: antral follicle count (AFC), total ovarian volume, ovarian stromal volume, MG value of ovary, volumetric ovarian vascular indices (VI, FI, VFI) and pulsed-wave Doppler indices of the main ovarian stromal vessels (PSV, EDV, RI, PI, CI). Assessment of measurement reliability was performed randomly on 10 cases (five control and five patients with PCOS) by calculating the intra-class correlation coefficient (ICC) and the 95% confidence interval (CI) by a one-way random effects model following three consecutive measurements for the 3D ultrasound parameters and two measurements for the pulsed-wave Doppler indices.

Results

Patient characteristics
We recruited 43 Caucasian women with PCOS as the study group. Three were excluded due to the presence of ovarian cysts (one with a dermoid cyst and two with endometriotic cysts), leaving a final study group of 40 patients. Of these 40 subjects, 10 (25%) were ovulatory hyperandrogenic women with PCO, 8 (20%) were anovulatory normoandrogenic women with PCO and the remaining 22 (55%) were anovulatory hyperandrogenic women with or without PCO. Among the 32 women with clinical and/or biochemical hyperandrogenism, 15 (46.9%) and 8 (25.0%) were moderately and severely hirsute while 7 (21.9%) and 3 (9.4%) suffered from moderate and severe acne, respectively.

We also recruited 42 subfertile Caucasian women without PCOS who were about to start IUI treatment in natural cycles, in association with OI, as the control group. Two of these subjects were excluded on the first ultrasound examination due to the presence of endometriotic cysts.

The mean BMI of PCOS group was significantly higher than the controls (27.35 ± 0.66 versus 24.1 ± 0.53 kg/m², P < 0.05) but there were no significant differences in the mean ages between groups (30.8 ± 0.8 versus 32.5 ± 0.5 years, P > 0.05).

Reliability of ultrasound measurements

The intra-observer reliability of 3-D volumetric & vascularity measurements was high with mean ICC ranging from 0.986 to 0.998. The measurements of vascular indices of pulsed-wave Doppler waveform also had high intra-observer reliability with
Ovarian stromal artery pulsed wave Doppler

Subject index; PI, pulsatility index; CI, capacitance index. PSV, peak systolic velocity; EDV, end-diastolic velocity; RI, resistance index.

Table I. The intra-observer reliability of ultrasound measurements of ovarian volume and ovarian vascular indices by power Doppler and pulsed-wave Doppler, as measured by the mean intra-class correlation coefficients (ICC) and their 95% confidence intervals (CI).

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean ICC</th>
<th>95% CI</th>
</tr>
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<tbody>
<tr>
<td>Ovary</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Antral follicle count</td>
<td>0.995</td>
<td>0.985–0.999</td>
</tr>
<tr>
<td>Total ovarian volume</td>
<td>0.998</td>
<td>0.993–0.999</td>
</tr>
<tr>
<td>Stromal volume</td>
<td>0.997</td>
<td>0.990–0.999</td>
</tr>
<tr>
<td>Mean grey value</td>
<td>0.987</td>
<td>0.962–0.996</td>
</tr>
<tr>
<td>VI</td>
<td>0.997</td>
<td>0.991–0.999</td>
</tr>
<tr>
<td>FI</td>
<td>0.988</td>
<td>0.966–0.997</td>
</tr>
<tr>
<td>VFI</td>
<td>0.997</td>
<td>0.993–0.999</td>
</tr>
<tr>
<td>Ovarian stromal artery</td>
<td></td>
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<tr>
<td>PSV</td>
<td>0.995</td>
<td>0.981–0.999</td>
</tr>
<tr>
<td>EDV</td>
<td>0.998</td>
<td>0.991–0.999</td>
</tr>
<tr>
<td>RI</td>
<td>0.986</td>
<td>0.949–0.997</td>
</tr>
<tr>
<td>PI</td>
<td>0.995</td>
<td>0.981–0.999</td>
</tr>
<tr>
<td>CI</td>
<td>0.994</td>
<td>0.975–0.999</td>
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</table>

VI, vascularization index; FI, flow index; VFI, vascularization flow index; PSV, peak systolic velocity; EDV, end-diastolic velocity; RI, resistance index; PI, pulsatility index; CI, capacitance index.

The ICC ranging from 0.987 to 0.999. The ICC and their 95% CI for each ultrasound parameter are summarized in Table I.

3D ultrasound features

The Kolmogorov–Smirnov test confirmed the data were not normally distributed. The data are presented as a median and range of values and the Mann–Whitney test was used to compare the differences between the groups. There were no differences in any of the ultrasound parameters between the right and left ovaries, so the mean values are presented and ovarian data per subject were analysed.

Comparisons between women with PCOS and their controls

Women with PCOS had significantly higher AFCs [median (range): 16.3 (9–35) versus 5.5 (2–10), \( P < 0.001 \)], total ovarian volume [12.56 (6.49–20.27) versus 5.66 (2.81–9.17) ml, \( P < 0.001 \)], ovarian stromal volume [10.79 (5.65–17.12) versus 4.69 (2.34–7.66) ml, \( P < 0.001 \)] and certain 3D measures of ovarian vascularization [VI: 3.85 (1.58–9.17) versus 2.79 (0.98–5.87), \( P < 0.001 \); VFI: 1.27 (0.42–4.11) versus 0.85 (0.30–2.27), \( P < 0.001 \)] than the controls. There were no significant differences in the ovarian Flow Index or MG value between the groups. There were also no significant differences in any measurements derived from analysis of the pulsed-wave Doppler waveform analysis between the groups. All of these results are summarized in Table II.

Subgroup analysis for women with variable manifestations of PCOS

In the subgroup analysis, 14 women with PCOS who were of normal weight (BMI < 25 kg/m²) had significantly higher ovarian echogenicity [MG, median (range): 32.68 (27.48–53.71) versus 28.00 (19.35–32.62), \( P < 0.05 \)] and ovarian vascular as measured by 3D ultrasound [VI: 4.51 (2.58–9.17) versus 3.25 (1.58–4.94), \( P < 0.05 \); and VFI: 1.56 (0.70–4.11) versus 1.22 (0.42–1.66), \( P < 0.05 \)] than their 11 obese counterparts (BMI ≥ 30 kg/m²). A group of 30 women with PCOS who were clinically hirsute also had significantly higher ovarian vascularity as measured by Flow Index [FI: 33.94 (26.39–51.65) versus 29.30 (21.88–36.78), \( P < 0.05 \)] than their 10 non-hirsute counterparts, but there were no significant differences in ovarian echogenicity. Hirsute women with PCOS had significantly increased stromal volume [11.48 (6.78–17.12) versus 9.68 (5.65–13.62) ml, \( P < 0.05 \)] compared with those non-hirsute women with PCOS. There were no significant differences in any of the ultrasound parameters between women with PCOS who were ovulatory and those who had chronic anovulation. Despite the noted differences in 3D measures of ovarian vascularity, there were no differences between the groups in any of the measurements derived from pulsed-wave Doppler waveform analysis. All of these results are summarized in Table III.

Discussion

PCOS is a complex heterogeneous endocrine disorder. The 2003 Rotterdam consensus represents an important first step...
Table III. 3D ultrasound features of PCOs in women with different phenotypic manifestations of PCOS compared with their counterparts.

<table>
<thead>
<tr>
<th>Subject</th>
<th>Parameter</th>
<th>Normal weight (n = 10)</th>
<th>Obese (n = 10)</th>
<th>Ovulatory (n = 10)</th>
<th>Anovulatory (n = 10)</th>
<th>Normo-androgenic (n = 10)</th>
<th>Hirsute (n = 10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ovary</td>
<td>Antral follicle count (n = 10)</td>
<td>16.0 (12–35)</td>
<td>15.5 (12–29)</td>
<td>15.0 (9–35)</td>
<td>16.0 (12–29)</td>
<td>15.5 (12–29)</td>
<td>17.3 (9–35)</td>
</tr>
<tr>
<td></td>
<td>Mean grey value</td>
<td>32.68* (27.48–53.71)</td>
<td>28.00* (19.35–32.62)</td>
<td>31.94 (24.74–40.74)</td>
<td>32.57 (19.35–53.71)</td>
<td>35.30 (27.48–53.71)</td>
<td>34.22 (25.51–51.65)</td>
</tr>
<tr>
<td></td>
<td>VFI (0–100)</td>
<td>1.56* (0.70–4.11)</td>
<td>1.22* (0.42–1.66)</td>
<td>1.31 (1.04–2.64)</td>
<td>1.26 (0.42–1.41)</td>
<td>1.07 (0.42–1.81)</td>
<td>1.30 (0.42–1.61)</td>
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<tr>
<td></td>
<td>Ovarian stromal artery pulsed wave Doppler</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>EDV (cm/s)</td>
<td>3.23 (2.18–5.19)</td>
<td>4.05 (1.54–6.35)</td>
<td>3.21 (2.04–4.43)</td>
<td>3.72 (1.54–7.44)</td>
<td>3.54 (2.18–5.22)</td>
<td>3.69 (1.54–7.44)</td>
</tr>
<tr>
<td></td>
<td>RI</td>
<td>0.65 (0.52–0.75)</td>
<td>0.59 (0.50–0.88)</td>
<td>0.61 (0.50–0.77)</td>
<td>0.61 (0.50–0.88)</td>
<td>0.60 (0.50–0.88)</td>
<td>0.60 (0.50–0.88)</td>
</tr>
<tr>
<td></td>
<td>PI</td>
<td>1.18 (0.85–2.22)</td>
<td>1.21 (0.85–2.17)</td>
<td>1.18 (0.85–2.22)</td>
<td>1.03 (0.85–2.22)</td>
<td>1.04 (0.85–2.22)</td>
<td>1.04 (0.85–2.22)</td>
</tr>
<tr>
<td></td>
<td>CI</td>
<td>2.08 (1.68–2.34)</td>
<td>2.03 (1.63–2.25)</td>
<td>2.09 (1.68–2.34)</td>
<td>2.03 (1.63–2.25)</td>
<td>2.05 (1.65–2.23)</td>
<td>2.03 (1.63–2.25)</td>
</tr>
</tbody>
</table>

Data given are presented as median (range). *p < 0.05 indicates significant differences within the group.

3D ultrasound allows for calculation of stromal volume, either manually or automatically using the inversion mode and thresholding. Our study is the first to calculate the ovarian stromal volume automatically using the latter technique (see Figs 1 and 2). We have shown stromal volume is significantly increased in women with PCO and more so in women with PCO who are clinically hirsute. This would support the hypothesis that the probable source of excessive androgen production in these groups of patients is the thecal cells in the ovarian stroma which undergo hypertrophy. Kyei-Mensah et al. also showed stromal volume was significantly higher in women with PCOS and PCO than in controls (16.7 and 15.0 ml versus 9.6 ml, all P < 0.05) (Kyei-Mensah et al., 1998). They examined 26 women with PCOS, 24 women with regular menstrual cycles but PCO on ultrasound scan and 50 subfertile women with regular menstrual cycles and normal ovarian morphology. They calculated the stromal volume through the subtraction of total follicular volume from total ovarian volume both of which were measured manually. Such measurements are time-consuming and more open to measurement error as a result. The fact that both techniques have shown a significant increase in stromal volume suggests some degree of compatibility and, while we cannot directly compare the time needed for the calculation of stromal volume by either technique, one must assume the automated technique is quicker.

Based on the Rotterdam diagnostic guideline, the hyperandrogenic criteria of PCOS can be fulfilled on a biochemical or clinical basis. Blood was not taken for serum androgen assay in our study as it aimed to evaluate the effects of different clinical presentations in women with PCOS on the 3D ultrasound features of PCOs. Clinical hyperandrogenism was assessed subjectively by patients on both hirsutism and acne aspects. Although the Ferriman Gallwey scoring system is the commonest instrument used to evaluate the medical
treatments for hirsutism, it has not been adopted in this study as we were not examining changes with treatment and the system has been shown to have poor interobserver agreement in women with PCOS (Wild et al., 2005). The majority (over 70%) of our hyperandrogenic cohort suffered from significant clinical hirsutism requiring regular shaving. We considered this an important complaint, i.e. in keeping with the Rotterdam consensus and therefore validates our omission of serum androgen levels, although these would have added value to the study.

The ovarian blood flow has repeatedly been shown to be increased in women with PCO using 2D ultrasound (Balen et al., 2003), and more recently with 3D ultrasound, although the studies using the latter modality have reported variable findings (Jarvela et al., 2002; Pan et al., 2002b; Ng et al., 2005). Pan et al., using a similar approach, also noted an increased ovarian volume and vascularity in 25 Chinese women with PCOS compared with 54 subfertile women with regular menstrual cycles and normal ovarian morphology (Pan et al., 2002b). Jarvela et al., however, concluded that there were no differences in ovarian vascularity between 14 women with PCO and a control group of 28 women (Jarvela et al., 2002), although the results must be interpreted with caution as the sample size was small and the group classifications unclear. Ng et al. reported no differences in ovarian blood flow in their study of 32 women with PCOS and 107 fertile controls (Ng et al., 2005). The lack of overall difference in ovarian vascularity between the groups may be explained by different patient populations. The study by Ng et al. included only anovulatory Chinese women with ultrasound appearances of PCO, and did not consider the degree of clinical or biochemical hyperandrogenism. Our study has shown there are no differences in women with anovulatory and ovulatory PCO and that any apparent differences relate more to body weight and biochemical hyperandrogenism with ovarian vascularity being more prominent in women with PCOS who are of normal weight, or who have hyperandrogenism. In keeping with this, Ng et al. did observe significantly higher ovarian blood flow in women with PCOS with a BMI <25 kg/m² than in their overweight counterparts. These findings suggest ovarian vascularity is influenced by the phenotypic expression of PCOS. Failure to assess, report or account for these differences may, in part, explain the variable results reported in the 3D studies to date, all of which have employed different study designs and reported different patient characteristics and therefore populations.

In this study, increased ovarian vascularity in women with PCO was only evident when measured by 3D power Doppler, with all 2D pulsed-wave Doppler measures showing no difference between the study populations or within the subgroups. Pulsed-wave Doppler has been used, however, to demonstrate differences in ovarian blood flow in women with PCOS in previous studies (Aleem and Predanic, 1996; Dolz et al., 1999; Zaidi et al., 1995). The lack of any identifiable difference with pulsed-wave Doppler in this study probably reflects the need to arbitrarily select a single vessel in the ovarian stroma rather than examine the total ovarian blood flow as occurs with 3D vascular quantification, which did reveal significant differences in these groups of women.

This is the first study to show differences in blood flow between any two groups that are only detectable by 3D ultrasound confirming the superiority of this technique. Interestingly, of the three different 3D indices of vascularity, only the VI and VFI were increased in the overall analysis comparing women with PCOS to controls. In contrast, the FI was significantly higher in women with PCOS who were hirsute compared with their normo-androgenic counterparts. These differences suggest the indices reflect different blood flow parameters, although we do not know how they truly equate to the actual vascularization and perfusion of the ovary.

The original diagnosis defined by Adams et al., 1985 included reference to an ‘increased stromal echogenicity’, but this was assessed subjectively which may explain its absence from the diagnostic criteria set by the Rotterdam consensus. 3D ultrasound allows for an objective measure of ovarian echogenicity through the calculation of the MG value which reflects the average intensity of the grey voxels within the VOI. Jarvela et al. are the only other group to have objectively examined stromal echogenicity as measured by the 3D calculation of the MG value of the whole ovary (Jarvela et al., 2002). They showed no differences in stromal echogenicity between 14 women with PCO and 28 women with ultrasonographically normal ovaries. Their classification of women with ‘PCO’ did not necessarily include those with clinical or endocrinological manifestations of PCOS which may have affected the power of the study and its ability to detect any real differences between the groups. Our study was based on the new Rotterdam diagnostic criteria and while our overall results concur with the findings by Jarvela et al. in that echogenicity is not higher in women with PCOS, we did observe a significantly higher ovarian echogenicity in women with PCOS who were of normal weight. This may reflect a more important pathogenic role of ovarian stroma in non-obese PCOS women who tend to have less metabolic disturbance (Ehrmann et al., 2006). Previous studies have looked at women with PCO as a single group and rarely assessed the effect of confounding variables such as weight, hirsutism or ovulation.

3D ultrasound is a relatively new imaging modality that permits improved spatial awareness, true volumetric calculation and quantitative assessment of the vascularity within a defined volume of tissue. This study revealed important differences between women with PCOS who are of normal weight or hyperandrogenic and their counterparts, supporting the concept that ovarian characteristics may influence, or be influenced by, the phenotypic expression of the disease. Increased stromal vascularity in women with PCOS who are of normal weight, or who are hirsute, suggests that ovarian stroma may play an important role in the development of hyperandrogenism. This study highlights the potential importance of the objective quantification of stromal echogenicity, ovarian and stromal volume and ovarian vascularity by 3D ultrasonography. Further prospective studies using these indices to predict treatment outcome are required to validate these findings and ascertain if they are of clinical value. Moreover, Rotterdam guidelines certainly form an important base for future studies on 3D ultrasound features of PCO as they ensure clinical standardization.
References


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