First trimester placental and myometrial blood perfusion measured by 3D power Doppler in normal and unfavourable outcome pregnancies

E. Hafner a,*, M. Metzenbauer a, I. Stümpfen a, T. Waldhör b, K. Philipp a

a Department OB Gyn, Geburtshilfliche Abteilung Donauspital am SMZ-Ost, Langobardenstraße 122, A–1220 Vienna, Austria
b Institute of Cancer Research, Department of Epidemiology, University of Vienna, Vienna, Austria

A R T I C L E   I N F O

Article history:
Accepted 17 June 2010

Keywords:
Placental perfusion
Myometrial perfusion
Uterine artery Doppler first and second trimester
Assessment of trophoblast invasion

A B S T R A C T

Introduction: To evaluate whether 3D placental and myometrial power Doppler blood perfusion in the first trimester can be used to detect risk pregnancies.

Methods: 3D power Doppler vascularization index (VI) and flow index (FI) of the entire placenta and the neighbouring myometrium were separately measured in the first trimester in all women with singleton pregnancies during a period of three months. In addition we measured placental volume, placental quotient, PAPP-A, as well as uterine artery at 12 and 22 weeks (mean PI and mean notch) and compared those data with the pregnancy outcome.

Results: Data from 383 women could be evaluated. 10 developed pre-eclampsia (PE). Both flow and vascularization were markedly lower in the placentas compared to the adjoining decidua and myometria. There was some correlation between placental vascularization index (PVI) as well as deciduo-myometrial vascularization index (MVI) and placental volume, PAPP-A and number of pregnancies and a marked reduction in PE-pregnancies (p: 0.0018, 0.0004). Of all measured parameters MVI showed the best sensitivity for the detection of PE.

Conclusion: The correlation between PVI and MVI in the first trimester and mean notch in the second trimester shows that they provide valuable information at as early as 12 weeks which normally so far is only available at 22 weeks by uterine artery Doppler flow. As MVI measures the percentage of vessels in the deciduo-myometrial area it could also provide information on trophoblast invasion. This hypothesis is supported in particular by a marked decrease of the MVI in pregnancy problems especially in PE-pregnancies.

© 2010 Elsevier Ltd. All rights reserved.

1. Introduction

The causes for pregnancy-associated problems like pre-eclampsia and intrauterine growth restriction are diverse and elusive, and yet, from a clinical point of view, there seems to be an underlying basis — namely abnormally low blood flow through the placenta [1–4]. A clinical example which supports this hypothesis is raised hope that women at risk could be treated prophylactically several weeks before the real disease starts, in order to avoid or at least mitigate any pregnancy problems. Nevertheless, it turned out that the detection of increased uterine artery impedance during the second trimester is only of limited use, as prophylactic treatment at this late stage is not sufficiently effective [11–14]. At best it may lead to better pregnancy surveillance. The next logical step was to perform uterine Doppler measurements in the first trimester to start a possible treatment at this early stage. Increased uterine artery impedance in the first trimester is unfortunately not as helpful as in the second trimester as its correlation with pregnancy problems is considerably weaker [15–18].

Uterine artery impedance reflects the resistance of blood flow in the decidual and myometrial spiral arteries of the mother. Fetal trophoblasts invade maternal tissue alongside these arteries, destroys their muscle fibres and thus remodelling these vessels into pliant channels, leading to low flow resistance. The activity of
this process is only inadequately captured by uterine artery Doppler impedance, as trophoblast invasion starts as early as the first trimester but Doppler studies show the best result many weeks afterwards at approximately 22–24 weeks.

A possible approach to assess trophoblast invasion in the first trimester, i.e. at the time when it actually takes place, could be to measure placental and myometrial vascularization and perfusion. Power Doppler sonography is a method which allows observation of the number and flow of small tissue-vessels. It has been stated that it could be superior to spectral Doppler in low velocity blood flow conditions [19,20]. 3D methods however can provide this information for the entire placenta and its adjoining myometrium. Some studies have focused on this particular aspect but they were mostly performed relatively late in pregnancy and the methods used were unconvincing as only parts of the placentas were measured [21–24].

In this study we present data of routine 3D power Doppler measurements of the entire placenta and the neighbouring myometrium, done between 11 and 14 weeks of gestation. We want to ascertain how placental and myometrial blood flow and vascularization behave at this early stage of pregnancy. By comparing these findings with second trimester uterine artery impedance and pregnancy outcome data we want to know whether these indices can be used to assess the trophoblast activity in order to detect risk pregnancies earlier in pregnancy.

2. Methods

All women who book for delivery in our hospital routinely receive a free nuchal translucency and “combined test” measurement between 11 and 14 weeks when they give their consent. At the same time we routinely measure the placental volume (PV) with 3D ultrasound and form the placental quotient (PQ = placental volume/crown-rump length) which has repeatedly been described [25–27]. We also examine both uterine arteries at the level of the internal cervical os and calculate the pulsatility index (PI) using the pulsed Doppler flow curves of five subsequent cardiac cycles, and assess the presence or absence of the so-called notching on both sides (notch was rated as present if an early diastolic incisure occurred in every flow cycle), enquire about smoking habits and measure body mass indices (BMI).

For this study all women with singleton pregnancies in the first trimester were prospectively enrolled during a period of three months. Apart from the routine procedures mentioned above, we also measured the power Doppler vascularization (VI) and flow index (FI).

To achieve this we used 3D Power Doppler volumes of the whole placenta and the adjoining myometrium in singleton pregnancies. The VI measures the number of colour voxels in a particular region of interest (ROI) in comparison to its grey voxels.

Fig. 1. Measurement of PVI and MVI. A: Placenta and myometrium with high vascularization and flow. 1: tracing of the placenta. Placenta is turned in a horizontal position, borders are traced. 2: tracing of the myometrium. If myometrium was thicker than 1 cm it was measured from its placental attachment up to 1 cm. If its thickness was less than 1 cm this smaller figure was taken for measurement. B: Placenta and myometrium with low vascularization and flow. 1: tracing of the placenta in the identical way as compared to A1 2: tracing of the myometrium.
In this way it registers the percentage of colour to grey voxels. The colour voxels themselves show different flow intensity leading to the PI which stands for the mean colour voxel intensity.

Due to repeatability reasons we tried to standardize the process of data acquisition as far as possible: To measure these indices in both placenta and myometrium a power Doppler colour box was placed over the entire placenta and the adjoining myometrium. The Doppler values were standardized at quality normal, the pulse repetition rate 0.9 KHz using a wall motion filter low1. The angle of the grey-scale picture was 70°, zoom 1.6, focus zone 1, X-beam CRI1, SRI3, OTI normal, harmonic frequency: high. Then a volume box of the same size was placed in exactly the same position as the colour box and the recording process was started using the maximum quality preset and finally stored. Recording was done with the same velocity in all women (10 s). All measurements were done with a Voluson 730 Expert (General Electric Company Fairfield Connecticut USA). In case of artefacts during volume recording due to fetal or bowel movements the recording process was repeated until a sufficient volume quality could be achieved.

All measurements were done by one operator (EH) in order to avoid inter-observer differences. The aforementioned indices were measured separately in placentas and myometrium.

- In order to calculate indices, placentas were rotated in a horizontal position in both A- and B-plane. Using “VOXEL”, an inbuilt programme for volume measurements, the placental border was carefully traced in the A-plane by calliper, which at the same time defines the region of interest (ROI). The placenta was then rotated by 30° in a horizontal plane and tracing of the placental border was repeated. The angle-size for the horizontal rotation was previously set. At an angle of 30° it takes six cuts to completely define the placental borders. After this, the machine calculates the VI and FI automatically (Fig. 1A1 and B1).
- The VI and FI in the myometrium were measured by the same method. Although placental borders are clearly defined, the exact extent of the myometrium is relatively imprecise, as its thickness differs considerably from a few millimetres to some centimetres. To solve this problem, the myometrium was measured from the direct attachment to the placenta up to a thickness of 1 cm which is possible by using the display-measure. If the thickness of the myometrium was less than 1 cm this figure was taken for measurement (Fig. 1A2 and B2). As this manual delineation might be a possible source of inaccuracy, measurements were performed twice on the same stored sample. The angle-size for the horizontal rotation was 70°. Zoom 1.6, focus zone 1, X-beam CRI1, SRI3, OTI normal, harmonic frequency: high.
- In order to find out whether indices differ according to placental position and its mean distance from the abdominal wall these data (anterior or posterior placental attachment, mean placental distance in cm from the abdominal surface) were collected in 75 women and the correlation calculated.

Women were then asked to return for a fetal anomaly scan at 22 weeks. At this stage we also measure the uterine artery PI and notch on both sides. For this purpose the uterine arteries are transabdominally identified by Colour flow mapping at the level where they cross the external iliac arteries. At least five pulsed Doppler cycles are recorded. Early diastolic notching is rated in the same way as the mean PI (PI right + PI left divided by two) and the mean notch (notch right + notch left divided by two, no notch = 1, notch = 2).

Pregnancy outcome data could be collected in all women who gave birth in our department. Women who gave birth in other hospitals in Vienna were followed up. So the following data were collected:

- Before birth:

### Table 1
Basic statistical data of 383 pregnant women.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Median</th>
<th>Min</th>
<th>Max</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>28.5</td>
<td>15</td>
<td>42</td>
</tr>
<tr>
<td>BMI</td>
<td>23.1</td>
<td>16.4</td>
<td>49.5</td>
</tr>
<tr>
<td>Length in cm</td>
<td>165.7</td>
<td>148</td>
<td>185</td>
</tr>
<tr>
<td>Gravidity</td>
<td>2</td>
<td>1</td>
<td>9</td>
</tr>
<tr>
<td>Parity</td>
<td>1</td>
<td>0</td>
<td>8</td>
</tr>
<tr>
<td>Cigarettes</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smokers</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>328</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non smokers</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>55</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

### Table 2
Correlations of the collected data to placental and myometrial indices. Correlations are significant at p < 0.05.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>PI</th>
<th>MFI</th>
<th>PVI</th>
<th>MVI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BMI</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Length in cm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gravidity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Parity</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cigarettes</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Smokers</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Author's personal copy

E. Hafner et al. / Placenta 31 (2010) 756–763
Table 3
Mean values and Variance of the measured parameters for different pregnancies and outcome problems.

<table>
<thead>
<tr>
<th></th>
<th>n</th>
<th>Mean PVI/ Std. Dev.</th>
<th>Mean MVI/ Std. Dev.</th>
<th>Mean PQ/ Std. Dev.</th>
<th>Mean PI at 12 weeks/ Std. Dev.</th>
<th>Mean notch at 12 weeks/ Std. Dev.</th>
<th>Mean PI at 22 weeks/ Std. Dev.</th>
<th>Mean notch at 22 weeks/ Std. Dev.</th>
<th>PAPP-A/ Std. Dev.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Normal outcome</td>
<td>307</td>
<td>9.79/5.88</td>
<td>32.31/12.38</td>
<td>0.94/0.22</td>
<td>1.81/0.62</td>
<td>1.65/0.41</td>
<td>0.95/0.28</td>
<td>1.12/0.29</td>
<td>1.10/0.64</td>
</tr>
<tr>
<td>SGA</td>
<td>41</td>
<td>8.27/5.39</td>
<td>30.40/11.32</td>
<td>0.71/0.17</td>
<td>1.94/0.70</td>
<td>1.70/0.42</td>
<td>1.02/0.32</td>
<td>1.20/0.37</td>
<td>1.05/0.44</td>
</tr>
<tr>
<td>Preterm delivery ≤ 34 weeks</td>
<td>13</td>
<td>7.46/4.23</td>
<td>29.09/13.36</td>
<td>0.89/0.35</td>
<td>1.85/0.53</td>
<td>1.58/0.45</td>
<td>0.85/0.24</td>
<td>1.08/0.28</td>
<td>1.02/0.77</td>
</tr>
<tr>
<td>Pre-pregnancy hypertension</td>
<td>5</td>
<td>5.56/3.14</td>
<td>27.49/8.81</td>
<td>0.77/0.15</td>
<td>1.97/0.50</td>
<td>1.90/0.22</td>
<td>1.28/0.54</td>
<td>1.30/0.44</td>
<td>1.10/0.58</td>
</tr>
<tr>
<td>PIH</td>
<td>7</td>
<td>6.03/3.29</td>
<td>21.97/11.02</td>
<td>0.91/0.14</td>
<td>1.74/0.54</td>
<td>1.71/0.39</td>
<td>1.02/0.34</td>
<td>1.43/0.53</td>
<td>1.41/0.67</td>
</tr>
<tr>
<td>PE</td>
<td>10</td>
<td>4.04/3.12</td>
<td>15.98/10.24</td>
<td>0.87/0.31</td>
<td>1.76/0.69</td>
<td>1.75/0.42</td>
<td>1.29/0.56</td>
<td>1.50/0.47</td>
<td>1.05/0.54</td>
</tr>
</tbody>
</table>

Fig. 2. Intra-observer difference in fifteen patients in the measurement of PVI and MVI using regression plots.

The guidelines of the International Society for the Study of Hypertension in Pregnancy (ISSHP) provided the basis for our classification of maternal complications, i.e. proteinuria, PIH and PE. Proteinuria was determined on two consecutive occasions on the basis of >2 or more proteinuria on the reagent strip urinalysis, or proteinuria of over 30 mg/24 h. PIH was judged as >140/90 mm Hg if recorded on two or more occasions during pregnancy, more than 155/105 on one occasion or the week at delivery, occurrence of pregnancy induced Hypertension (PIH) or Pre-eclampsia (PE), mode of delivery.

Fetal and placental data: crown-rump length (CRL), PV, PQ, placental vascular index (PVI), placental flow index (PFI), myometrial vascular index (MVI), myometrial flow index (MFI), free β h-CG, PAPP-A.

Maternal data: Age, gravidity, parity, BMI, history of high blood pressure, length in cm, cigarette smoking, mean PI at 12 weeks, mean notch at 12 weeks, mean PI at 22 weeks, mean notch at 22 weeks.
need for antihypertensive therapy during the pregnancy. PE was defined as a condition of PIH and proteinuria.

Women who did not show up at 22 weeks for the uterine Doppler scan were excluded from calculation. Pregnancies with fetal aneuploidies were also excluded. Uterine bleeding however was not considered as a reason for exclusion.

3. Statistics

Associations were calculated by the non-parametric Spearman’s correlation coefficient in SAS (SAS/STAT User’s Guide, Version 9, Cary, NC 27513: SAS Institute Inc. 2002–2003). A group consisting of PE and PIH-pregnancies was formed and compared with the group of normal outcome pregnancies using the Kruskal–Wallis test. Pair-wise comparisons for all measured parameters between normal versus PE and PIH-pregnancies were done by the Wilcoxon test. P-values for those pair-wise comparisons were adjusted by the step-down Bonferroni option in SAS (procedure multtest). ROC-curves were calculated based on logistic regression models.

4. Results

Placental and myometrial vascularization and flow in 423 singleton pregnancies were measured. Six had to be excluded due to fetal aneuploidies or malformations. Four women suffered from abortion between 12 and 22 weeks. 30 women did not show up at the scheduled time of fetal anomaly scan for unknown reasons or pregnancy outcome could not be followed up, which left a total of 383 women for evaluation.

Maternal data are shown in Table 1.

The correlations of the collected data to placental and myometrial indices can be seen in Table 2. The number of pregnancies and births has some influence on both flow and vascularization parameters. CRL has no significant influence on placental or myometrial indices, except for a weak positive correlation with MVI. Correlations between flow and vascularization parameters and PV and PQ are also weak, indicating that the number of vessels as well as the amount of flow hardly increases during the observed period of time.

The strongest correlation can be seen between MVI and mean uterine artery notch at 22 weeks (Correlation Coefficient mean MVI and mean notch 22 weeks: –0.322). This correlation is stronger than at 12 weeks (Correlation Coefficient mean MVI and mean notch 12 weeks: –0.302). The same is true with mean MVI and mean PI at 22 and 12 weeks.

The repeatedly published correlation between PAPP-A and PV/PQ can also be observed in our present data. The correlation between PAPP-A and both PVI and MVI is comparably weaker and, with regard to flow indices, does not exist at all which could indicate that placental or myometrial perfusion is not deeply involved in the production of this hormone.

Contrary to our expectations, cigarette smoking did not show any influence on placental or myometrial flow and vascularization.

There was no correlation between any of the indices examined (PFI, MFI, PVI and MVI) and placental attachment (anterior vs. posterior placenta). There was also no correlation between PI and MVI and mean placental distance from the abdominal surface. PFI and MFI however showed a considerable correlation to the mean placental distance from the abdominal surface (Spearman Correlation Coefficient for PFI: –0.308 and for MFI: –0.336), indicating that flow indices in contrast to vascularization indices are strongly depth dependent and hence less reliable. Maternal BMI consequently has a significant negative influence on flow indices but only a slight negative influence on vascularization indices. This suggests that flow indices are relatively unreliable in obese women, whereas vascularization parameters are nearly independent from maternal obesity (correlation coefficient PFI and BMI: –0.133, MFI and BMI: –0.239, MVI and BMI: –0.11, PVI and BMI: ns).

![Flow and vascularization differences in placenta and myometrium](image-url)
Author's personal copy

For this reason and because of the more reliable relationship between VI and true flow-characteristics mentioned above [30–32], we only used the PVI and MVI for the evaluation of the pregnancy outcome.

Table 3 shows the mean values of the measured parameters for normal outcomes in comparison to different outcome problems. The group of PE and PIH-pregnancies showed a significant difference compared to the group of normal pregnancies for the PVI, MVI, mean PI at 22 weeks and mean notch at 22 weeks respectively (p-value: 0.0007, 0.0001, 0.0367, 0.0019), but no difference for mean PI at 12 weeks, mean notch at 12 weeks, PQ, PAPP-A. The pair-wise comparison between normal and PIH-pregnancies was significantly different for the MVI and mean notch at 22-week stage (p-values: 0.0393, 0.0162). The pair-wise comparison between normal and PE-pregnancies was significantly different for PVI, MVI, mean PI at 22 weeks and mean notch at 22 weeks (p-values: 0.0018, 0.0004, 0.028, 0.016). Pair-wise comparisons for all other parameters did not show statistically significant differences.

Fig. 1 shows the method of tracing and measuring placental and myometrial indices.

Fig. 2 shows a regression plot for intra-observer differences. The adjusted R-square-values explain the percentage of the variance for the measured values. This was 0.7913% for the PVI and 0.981% for the MVI (range 0–100%) indicating that it is especially the MVI which can most reliably be measured.

Fig. 3 shows that median placental indices are markedly lower than myometrial and the range smaller. Vascularization indices differ more than flow indices (Median PFI: 43.69, range 35.22, median MFI: 54.89, range 73.58, median PVI: 8.40, range 29.56, median MVI: 30.98, range 73.58).

Fig. 4 shows ROC-Curves for the detection of PE using different parameters (PVI, MVI, mean PI at 12 weeks and mean PI at 22 weeks), indicating that MVI is slightly superior to PVI, mean PI at 22 weeks and especially at 12 weeks.

5. Discussion

Many attempts have been made to find first trimester parameters which are able to detect pregnancies at risk for IUGR and PE. According to a common hypothesis, these severe problems are a consequence of low placental blood flow due to impaired trophoblast invasion [1–4]. Efforts have been made to assess reduced placental blood flow using uterine artery spectral Doppler. Large scale studies, however, show that the sensitivity of this method is limited if used as a screening tool in a low-risk population [34–37]. The reason for this might be that uterine artery impedance depends on a great number of factors including the initial size of the uterus, gravidity, maternal length, placental volume, trophoblast activity etc. Hence, differences in uterine artery impedance are not exclusively caused by spiral artery remodelling, but also depend on other factors, which may lead to variable growth, adaptation and velocity, until the final end-point — low impedance and no notching — is achieved. Only then — at approximately 24 weeks — can reliable evidence be obtained and conclusions drawn. With the 3D power Doppler method presented in this study, the result of trophoblast invasion, i.e. remodelling of spiral vessels and subsequent increase in vascularization, could possibly be measured directly.

This assumption is strengthened by the fact that we found the strongest correlation between MVI and mean uterine artery impedance at 22 weeks. This correlation was better at 22 weeks as compared to 12 weeks. This finding shows that some of the information which normally can only be obtained later on by the presence of a notch and high uterine PI in the second trimester is already available with the MVI in the first trimester.
Our findings show that the decidua and myometrium are much better perfused than the placenta itself. Placental indices are much lower and have a smaller range than myometrial indices. The really interesting part of the perfusion takes place in the decidua and myometrium. The MVI provides direct insights into the number of vessels in this particular region which enables the activity of trophoblast invasion to be assessed. This hypothesis is supported by the great differences in PVI/MVI in pregnancies with outcome problems. Differences between normal and SGA-pregnancies are only slight because most of the small newborns are genetically small. Their placentas are already small at 12 weeks (low PQ) but well perfused. The group of hypertensive pregnancies, however, shows a significantly reduced perfusion at 12 weeks, which is especially true for preeclamptic pregnancies. The explanation for this is that these problems are caused mainly by shallow trophoblast invasion and its consequences.

A moot point in 3D Power Doppler is its repeatability. It has been shown that VI and FI are significantly influenced by power Doppler settings like gain, signal power and pulse repetition frequency. They are also affected by volume flow, attenuation, vessel number and erythrocyte density, but VI in a linear and FI in a cubic way. There was a marked reduction in each index as the distance between the transducer and vessel increased, with a linear relationship between depth and VI and a more complex relationship to the FI especially above 55 mm. So the VI seems to have a more predictable relationship whereas the FI demonstrates a complex cubic relationship [30–32]. To reduce these problems it is important to point out that the preset of the ultrasound machine used must always be identical as changes in grey values and especially colour parameters like pulse repetition rate and high pass filter lead to substantial differences in the number of colour voxels compared to grey voxels, or the assessment of colour intensity. In other words, different settings on the ultrasound machine lead to different results. When we measured the vascularization or flow indices in the same woman with two different sonographical presets, results could differ substantially. Power Doppler is depth dependent [30–33]. Our data show that there is no correlation with placental attachment but a negative correlation of the vascularization to outcome data, however, shows that especially MVI is promising with respect to risk pregnancies. We think this justifies this method to be employed.

In a similar way to the success in the detection of Down’s syndrome using a combination of sonographical and biochemical markers, it will be interesting to combine the vascularization data with substances that have proven to be effective for the detection of PE in the first trimester. Studies have been published using a combination of uterine artery Doppler at 12 weeks with PAPP-A and PP13. Although the results were promising they were not convincing enough to be established as a screening tool in a low-risk population [17,38]. The main cause is probably the relative weakness of first trimester uterine artery Doppler. It would be of interest whether MVI is able to substantially improve sensitivity in similar combinations, or how different excursions from some of the biochemical markers in question influence placental and myometrial vascularization and flow.

References


