



## ISUOG Practice Guidelines (updated): use of Doppler velocimetry in obstetrics

### Clinical Standards Committee

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### SCOPE OF THE DOCUMENT

This document is a Practice Guideline on how to perform Doppler ultrasonography of the fetoplacental circulation. It is of the utmost importance not to expose the embryo or fetus to unduly harmful ultrasound energy, particularly in the earliest stages of pregnancy. At these stages, Doppler recording, when clinically indicated, should be performed at the lowest possible energy levels. ISUOG has published guidance on the use of Doppler ultrasound at the 11 to 13+6-week fetal ultrasound examination<sup>1</sup>. When performing Doppler imaging, the displayed thermal index should be  $\leq 1.0$  and the exposure time should be kept as short as possible, usually no longer than 5–10 min.

It is not the intention of this Guideline to define clinical indications, specify appropriate timing of Doppler examination in pregnancy or discuss how to interpret findings or the use of Doppler in fetal echocardiography. The aim is to describe pulsed Doppler ultrasound and

its different modalities: spectral Doppler, color flow mapping and power Doppler, which are commonly used to study the maternal–fetal circulation. We do not describe the continuous-wave Doppler technique, because this is not usually applied in obstetric imaging; however, in cases in which the fetus has a condition leading to very high-velocity blood flow (e.g. aortic stenosis or tricuspid regurgitation), it might be helpful in order to define clearly the maximum velocities by avoiding aliasing.

The techniques and practices described in this Guideline have been selected to minimize measurement error and improve reproducibility. They may not be applicable in certain clinical conditions or for research protocols.

Details of the grades of recommendation used in this Guideline are provided in Appendix 1. Reporting of levels of evidence is not applicable to this Guideline.

### RECOMMENDATIONS

#### What equipment is needed for Doppler evaluation of the fetoplacental circulation?

- Equipment should have color flow and spectral wave Doppler capabilities, with onscreen display of flow velocity scales or pulse repetition frequency (PRF) and Doppler ultrasound frequency (in MHz).
- The mechanical index (MI) and thermal index (TI) should be displayed on the ultrasound screen and the ALARA (as low as reasonably achievable) principle should be applied during examination to ensure safety. **(GOOD PRACTICE POINT)**
- The ultrasound system should generate a maximum velocity envelope (MVE) showing the whole spectral Doppler waveform.
- It should be possible to delineate the MVE using automatic or manual waveform traces.
- The system software should be able to estimate peak systolic velocity (PSV), end-diastolic velocity (EDV) and time-averaged maximum velocity (TAMX) from the MVE and to calculate the commonly used Doppler indices, i.e. pulsatility (PI) and resistance (RI) indices and the peak systolic/end-diastolic velocity ratio (S/D ratio). The spectral trace should indicate the various

points included in the calculation of the Doppler indices. (GOOD PRACTICE POINT)

### Basic technical aspects

All Doppler modalities are based on three fundamental principles. (1) Moving structures change the frequency and amplitude of reflected ultrasound signals. Moving structures include not only blood, but also fetal vessels or tissues. This can generate a shift in the backscattered signals. (2) Analysis of the components of the reflected signals are utilized for different Doppler modalities: the shift in frequency for directional color and spectral Doppler, and the shift in amplitude for power Doppler ultrasound (PDU). (3) All color and power Doppler modalities are pulsed techniques, while spectral Doppler can be pulsed or continuous.

PRF, or scale, is the frequency at which the ultrasound signals (pulses) are emitted; a low PRF allows signals from slow-moving targets to reach the transducer before the next pulse is emitted, whereas a high PRF will allow only high velocities to reach the ultrasound transducer before the next pulse. The wall filter is a barrier defined by a specific threshold frequency below which signals are not displayed in the Doppler image. Gain is the amplification of signals. The quality and reproducibility of the recordings can be improved by knowledge of these Doppler settings and how to adjust them.

### How can the acquisition of Doppler waveforms be optimized?

#### *Spectral pulsed-wave Doppler ultrasonography*

- Recordings should be obtained in the absence of fetal breathing and body movements and, if necessary, during temporary maternal breath-holding.
- Color flow mapping is not mandatory, although it is very helpful in identification of the vessel of interest and in defining the direction of blood flow.
- The optimal insonation is completely aligned with the direction of blood flow. This ensures the best conditions for assessing absolute velocities and waveforms. Small deviations in angle may occur. An insonation angle of 10° corresponds to a 2% error in the velocity, whilst an angle of 20° corresponds to 6% error. When absolute velocity is the clinically important parameter (e.g. for the middle cerebral artery (MCA)) and an angle of close to 0° cannot be obtained, despite repeated attempts, angle correction may be used. In this case, a statement should be added to any report, noting the angle of insonation and whether angle correction was carried out or that the uncorrected velocity is recorded.
- It is advisable to start with a relatively wide Doppler gate (sample volume) to ensure the recording of maximum velocities during the entire pulse. If interference from other vessels causes problems, the gate can be reduced to refine the recording. It should be kept in mind that the sample volume can be reduced only in height, not in width.

- Similar to grayscale imaging, the penetration and resolution of the Doppler beam can be optimized by adjusting the frequency (in MHz) of the Doppler transducer.
- The vessel wall filter, variously called 'low-velocity reject', 'wall-motion filter' or 'high-pass filter', is used to eliminate noise resulting from the movement of the vessel walls. According to convention, it should be set as low as possible ( $\leq 50$ –60 Hz), in order to eliminate the low-frequency noise from peripheral blood vessels. When using a higher threshold for the filter, a gap between the Doppler line and the Doppler signals can be seen. This can create the spurious effect of absent EDV (see Figure 4b).
- A higher wall filter is useful to obtain a well-defined MVE from structures such as the aortic and pulmonary outflow tracts, which have high-velocity flows. A lower wall filter might cause noise, appearing as flow artifacts close to the baseline or after valve closure.
- Doppler horizontal sweep speed should be fast enough to separate successive waveforms. Ideal is a display of four to six (but no more than eight to 10) complete cardiac cycles. For fetal heart rates of 110–150 bpm, a sweep speed of 50–100 mm/s is adequate. (GOOD PRACTICE POINT)
- The PRF should be adjusted according to the vessel studied: a low PRF will enable visualization and accurate measurement of low-velocity flow; however, it will produce aliasing when high velocities are encountered. The waveform should fill at least 75% of the Doppler screen (see Figure 3). (GOOD PRACTICE POINT)
- Doppler measurements should be reproducible; therefore, it is recommended to obtain more than one Doppler recording. If there is an obvious discrepancy between two measurements, another recording is recommended. The most technically superior recording (which usually means the one with highest MVE) should be used for reporting. (GOOD PRACTICE POINT)
- Most ultrasound systems display the average of the indices of three consecutive waveforms obtained from each Doppler recording.
- In order to increase the quality of Doppler recordings, frequent updates of the real-time grayscale or color Doppler image should be performed (i.e. after confirming in the real-time image that the Doppler gate is positioned correctly, the two-dimensional (2D) grayscale and/or color Doppler image should be frozen while the Doppler waveforms are being recorded).
- Correct positioning and optimization of the Doppler recording of the frozen 2D image should be ensured by listening to the audible representation of the Doppler shift over the speaker.
- Simultaneous use of grayscale, color flow mapping and spectral Doppler (triplex mode) significantly negatively affects the quality of acquired data and is discouraged.
- Doppler gain should be adjusted in order to see clearly the Doppler velocity waveform, without the presence of artifacts in the background of the display.



- It is advisable not to invert the Doppler display on the ultrasound screen. In the evaluation of the fetal heart and central vessels, it is very important to maintain the original direction of the color flow and pulsed-wave Doppler display. Conventionally, flow towards the ultrasound transducer is displayed as red and the waveforms are above the baseline, whereas flow away from the transducer is displayed as blue and the waveforms are below the baseline. (GOOD PRACTICE POINT)

#### Color directional Doppler ultrasonography

- Compared with grayscale imaging, color Doppler increases the total power emitted. Color Doppler resolution increases when the color box is reduced in size. Care must be taken in assessing the MI and TI as they change according to the size and depth of the color box.
- Increasing the size of the color box also increases the processing time and thus reduces the frame rate; the box should be kept as small as possible, to include only the studied area.
- The velocity scale or PRF should be adjusted to represent the blood flow velocities of the studied vessel. When the PRF is high, low-velocity vessels will not be displayed on the screen. When a low PRF is applied incorrectly, aliasing will present as contradictory color velocity codes and ambiguous flow direction.
- As for grayscale imaging, color Doppler resolution and penetration depend on the ultrasound frequency. The frequency for the color Doppler mode should be adjusted to optimize the signals.
- Gain should be adjusted in order to prevent noise and artifacts, seen as a random display of color dots in the background of the screen.
- The filter should also be adjusted to exclude noise from the region studied.
- The angle of insonation affects the color Doppler image; it should be adjusted by optimizing the position of the ultrasound probe according to the vessel or area studied.

#### Power Doppler and directional power Doppler ultrasonography

- The same fundamental principles as those for color directional Doppler ultrasonography apply to PDU and directional PDU.
- PDU is more sensitive to lower velocities than is color directional Doppler.
- PDU detects changes in the amplitude (power) of Doppler signals.
- The angle of insonation has less effect on power Doppler than it does on color Doppler signals; nevertheless, the same optimization process as for color directional Doppler should be performed.

- There is no aliasing phenomenon using PDU (except directional PDU); however, an inappropriately low PRF may lead to noise and artifacts.
- Gain should be reduced in order to prevent amplification of noise (seen as uniform color in the background). PDU persistence should also be adjusted; high persistence adds PDU information to that from a previous image, while no persistence shows the dynamic changes in PDU in each frame. High persistence is useful when the vascularity of an area is evaluated.

#### What is the appropriate technique for obtaining uterine artery Doppler waveforms?

Using real-time color Doppler ultrasound, the main branch of the uterine artery is located easily at the cervicocorporeal junction. Doppler velocimetry measurements are usually performed near to this location, either transabdominally<sup>2</sup> or transvaginally<sup>3-5</sup>. While absolute velocities are of little or no clinical importance, semiquantitative assessment of the velocity waveforms is commonly employed. Measurements should be reported independently for the right and left uterine arteries, and the presence of notching should be noted. (GOOD PRACTICE POINT)

Notching is defined qualitatively as reduced early diastolic velocities before the maximum diastolic velocity in the Doppler waveform. The severity of notching is defined by the difference between the lower early and the maximum diastolic velocities<sup>6</sup>.

#### First-trimester uterine artery evaluation (Figure 1)

##### 1. Transabdominal technique

- Transabdominally, a mid-sagittal section of the uterus is obtained, and the cervical canal is identified.
- The probe is then moved laterally until the paracervical vascular plexus is seen.
- Color Doppler is turned on and the uterine artery is identified as it turns cranially, to make its ascent to the uterine body.

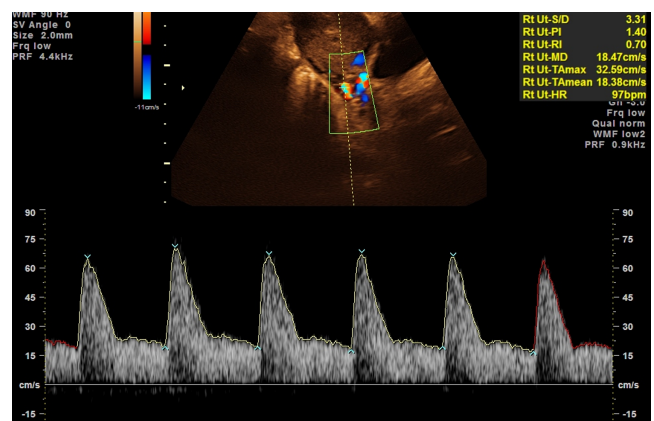


Figure 1 Waveform from uterine artery obtained transabdominally in first trimester.

- Measurements are taken at the point before the uterine artery branches into the arcuate arteries.
- As the PSV decreases from the uterine to the arcuate arteries, a measurement of PSV < 5<sup>th</sup> centile (60 cm/s)<sup>7</sup> should prompt the operator to verify carefully the placement of the sample volume.

The same process is repeated on the contralateral side. An alternative approach to obtain the Doppler signals using a cross-sectional plane has been described<sup>8</sup>, and showed comparable values and equally good reproducibility when compared to the sagittal plane<sup>9,10</sup>.

## 2. Transvaginal technique

- The woman should be asked to empty her bladder and should be placed in the dorsal lithotomy position.
- Transvaginally, the probe is placed in the anterior fornix. Similar to the transabdominal technique, the probe is moved laterally to visualize the paracervical vascular plexus, and the same steps are carried out in the same sequence as for the transabdominal technique.
- Care should be taken not to insonate the cervicovaginal artery (which runs in a cranial to caudal direction) or the arcuate arteries.

## Second- and third-trimester uterine artery evaluation (Figure 2)

### 1. Transabdominal technique

- Transabdominally, the probe is placed longitudinally in the lower lateral quadrant of the abdomen, angled medially in the parasagittal plane. Color flow mapping is useful to identify the uterine artery as it is seen crossing the external iliac artery.
- The uterine arteries usually run along each side of the uterus towards the fundus. To obtain the best angle of insonation, the position of the probe should be adjusted according to the orientation of the uterine artery.
- The sample volume is placed 1 cm downstream from this crossover point.
- In a small proportion of cases, the uterine artery branches before the intersection of the external iliac artery. In such cases, the sample volume should be placed on the uterine artery just before its bifurcation.
- The same process is repeated for the contralateral uterine artery.
- With advancing gestational age, the uterus usually undergoes dextrorotation. Thus, the left uterine artery does not run as lateral relative to the uterus as does the right.

### 2. Transvaginal technique

- The woman should be asked to empty her bladder and should be placed in the dorsal lithotomy position.
- The probe is placed in the lateral fornix and the uterine artery identified, using color Doppler, at the level of the internal cervical os.

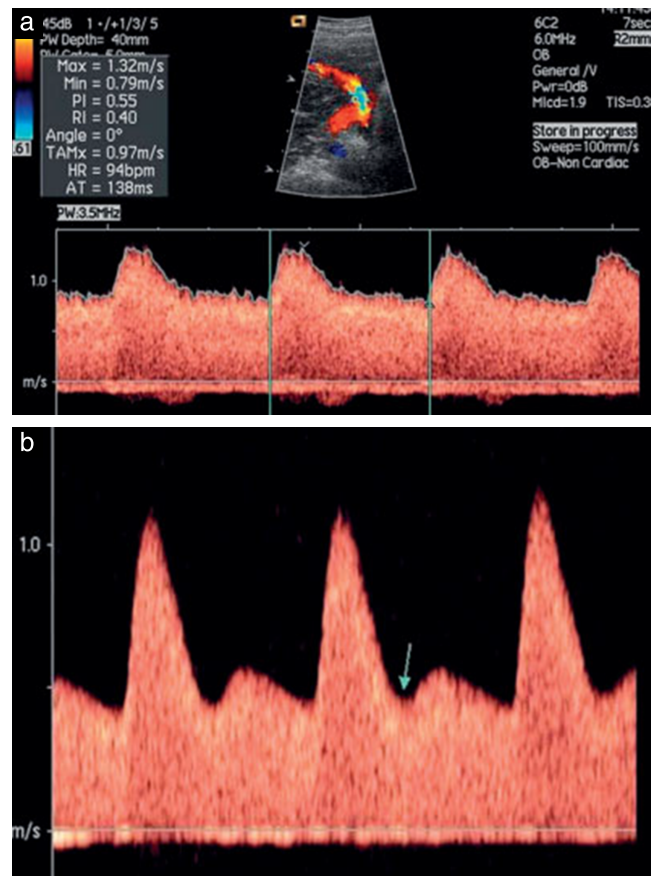


Figure 2 Waveforms from uterine artery obtained transabdominally in second trimester. Normal (a) and abnormal (b) waveforms; note notch (arrow) in Doppler signal in (b).

- This should then be repeated for the contralateral uterine artery. It should be remembered that reference ranges for uterine artery Doppler indices depend on the technique of measurement, so appropriate corresponding reference ranges should be used for transabdominal<sup>3</sup> and transvaginal<sup>5</sup> routes. The insonation technique used should be the same as that used for establishing the reference range.

Note that, in women with congenital uterine anomaly, assessment of uterine artery Doppler indices and their interpretation is unreliable, since all published studies have been on women with (presumed) normal anatomy. (GOOD PRACTICE POINT)

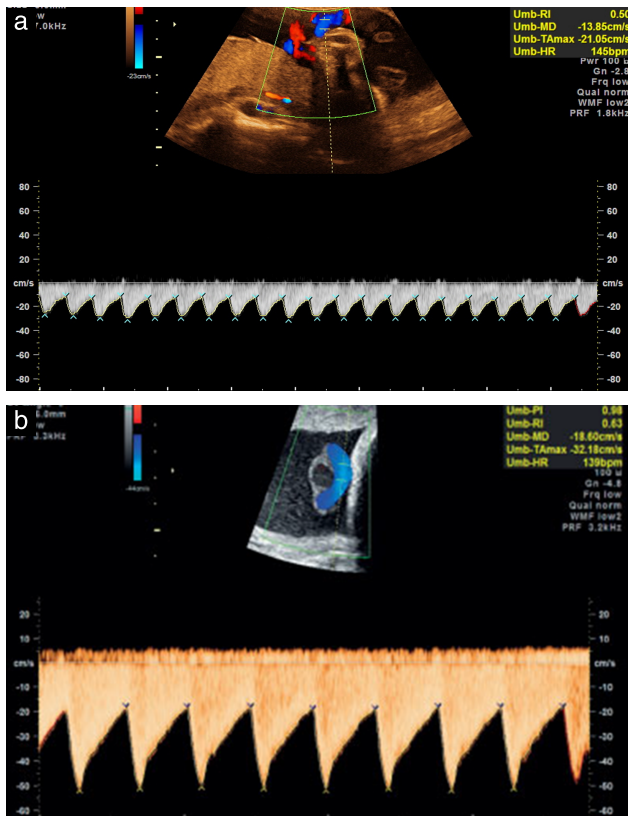
## What is the appropriate technique for obtaining umbilical artery Doppler waveforms?

There is a significant difference in Doppler indices measured at the fetal end (intra-abdominal)<sup>11</sup>, in a free loop and at the placental end of the umbilical cord<sup>12</sup>. The impedance is highest at the fetal end, and absent/reversed EDV is likely to be seen first at this site. Reference ranges for umbilical artery Doppler indices at each of these sites have been published<sup>11,13</sup>. For the sake of simplicity and consistency, by convention, measurements should be made in a free cord loop. (GOOD PRACTICE POINT)

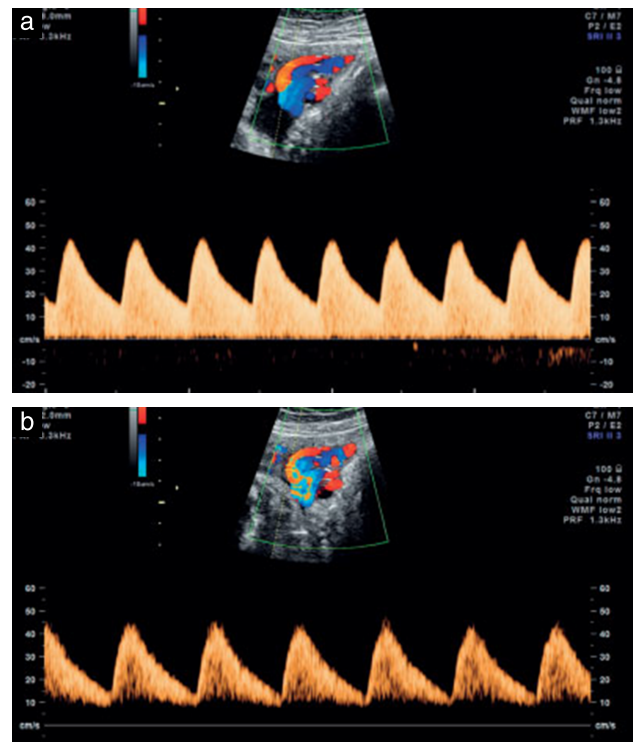
The decision to use a free loop of the cord was made early in the history of Doppler ultrasound and has been applied with great clinical success. However, in multiple pregnancies, and/or when comparing repeated measurements longitudinally, recordings from fixed sites, i.e. fetal end, placental end or intra-abdominal portion, may be more reliable. Appropriate reference ranges should be used according to the site of interrogation. Figure 3 shows examples of acceptable and unacceptable velocity waveform recordings and Figure 4 illustrates the influence of the vessel wall filter.

Note that, in multiple pregnancy, assessment of umbilical artery blood flow can be challenging, since there may be difficulty in assigning a cord loop to a particular fetus. It is therefore better to sample the umbilical artery just distal to the abdominal insertion of the umbilical cord. However, the impedance there is higher than that in a free loop and that at the placental cord insertion, so appropriate reference charts are needed. (GOOD PRACTICE POINT)

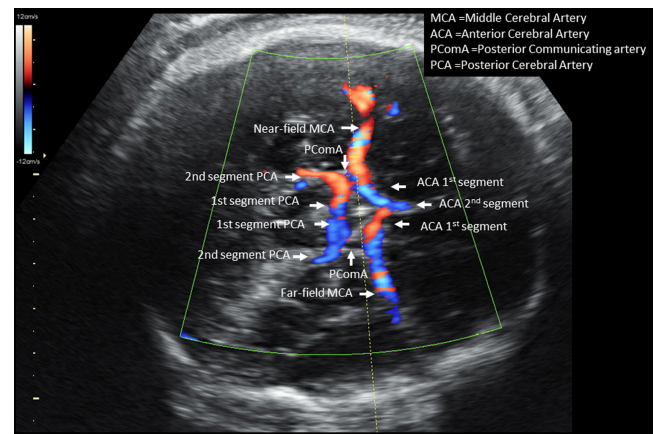
Note also that, in a two-vessel cord, at any gestational age, the diameter of the single umbilical artery is larger than the arterial diameter would be if there were two arteries<sup>14</sup>. Due to the different hemodynamics, the recorded velocity waveform in such cases should be interpreted with caution when using conventional reference ranges. (GOOD PRACTICE POINT)



**Figure 3** Examples of unacceptable (a) and acceptable (b) umbilical artery waveforms. The recording is improved by reducing the Doppler scale (i.e. reducing the pulse repetition frequency) to magnify the velocity recording on the screen, as well as adjusting the sweep speed to cover only three to nine consecutive waves.



**Figure 4** (a) Umbilical artery velocity waveform recorded with a low vessel wall filter setting showing normal flow and (b) a recording with apparently very low diastolic flow and absent flow signals at baseline, due to use of incorrect vessel wall filter, which is set too high, thereby concealing the low velocities along the zero line.



**Figure 5** Color flow mapping of the circle of Willis.

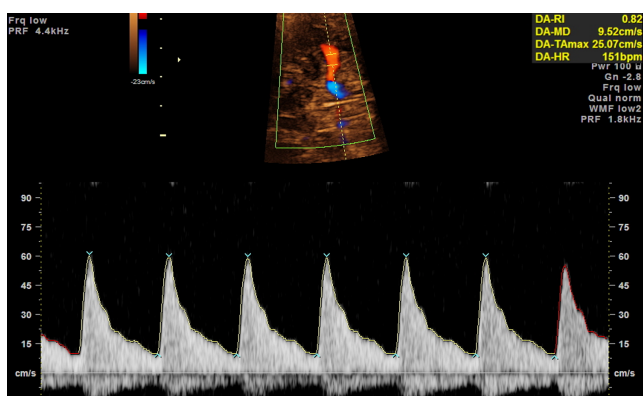
**What is the appropriate technique for obtaining fetal middle cerebral artery Doppler waveforms?**

- An axial section of the brain, including the thalami and the sphenoid bone wings, should be obtained and magnified.
- Color flow mapping should be used to identify the circle of Willis and the proximal MCA, just caudal to the transthalamic plane (Figure 5).
- The pulsed-wave Doppler gate should then be placed at the proximal third of the MCA, close to its origin in the

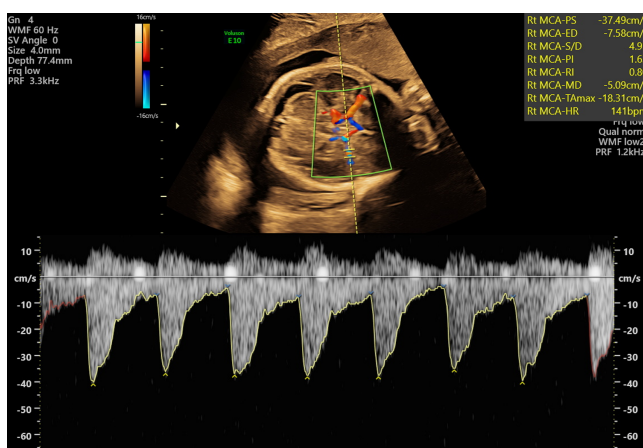


internal carotid artery<sup>15</sup> (the systolic velocity decreases with increasing distance from the point of origin of this vessel). (GRADE OF RECOMMENDATION: C)

- The angle between the ultrasound beam and the direction of blood flow should be kept as close as possible to 0° (Figure 6).
- Care should be taken to avoid any unnecessary pressure on the fetal head, as this may lead to increased PSV, decreased EDV and increased PI<sup>16</sup>.
- At least three and fewer than 10 consecutive waveforms should be recorded. The highest point of the waveform is considered as the PSV (in cm/s).
- The PSV can be measured using manual calipers or autotrace. PI is commonly reported using autotrace measurement, but manual tracing is also acceptable. In fact, manual caliper placement was used in the seminal work investigating the value of MCA-PSV for non-invasive detection of fetal anemia<sup>15</sup>.
- Appropriate reference ranges should be used for interpretation, and the measurement technique should be the same as that used to construct the reference ranges.



**Figure 6** Acceptable Doppler waveform from the middle cerebral artery. Note the angle of insonation has been adjusted to be nearly 0°.



**Figure 7** Middle cerebral artery (MCA) Doppler waveforms obtained from the far-field MCA. Note the 0° angle of insonation.

- The interobserver reliability of MCA-PI measurement is reported to be only moderate, with limited agreement between two observers. The 95% interval of the PI differences between observers was +0.91 to -1.14 at the proximal sampling site of the near-field MCA. In about 30% of cases, the PI difference between observers was greater than 0.5<sup>17</sup>. Multiple measurements are recommended to assess the true value.
- MCA-PSV measurements at the proximal site of the MCA in the near field are comparable to those obtained from the far-field vessel in clinical practice<sup>18,19</sup>. The far-field vessel may be chosen if obtaining an insonation angle of 0° is easier for the far-field than for the near-field MCA (Figure 7). (GRADE OF RECOMMENDATION: C)

### How are the cerebroplacental ratio and the umbilicocerebral ratio calculated?

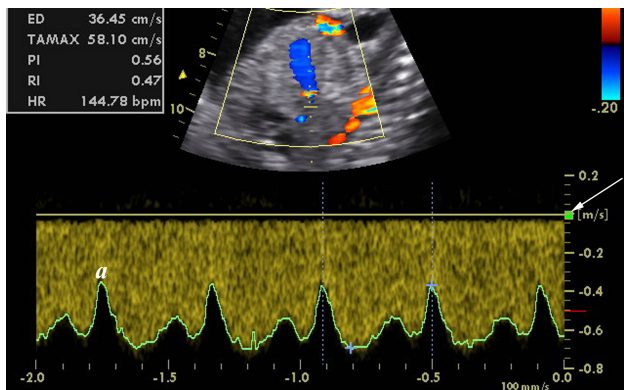
- The physiological basis for clinical application of the cerebroplacental Doppler ratio (CPR) is two-fold. The CPR is a reflection of the arterial redistribution that occurs during preferential brain perfusion in response to fetal hypoxemia ('brain sparing'). It amplifies mathematically the effect of abnormal hemodynamics in the umbilical and cerebral circulations and correlates more closely with the fetal partial pressure of oxygen (pO<sub>2</sub>) than does either of its component indices<sup>20-22</sup>.
- The ratio of Doppler indices from the cerebral and umbilical arterial circulations has been calculated variously, using indices of the MCA, anterior cerebral artery, vertebral artery or internal carotid artery, using umbilical artery indices in the denominator rather than the numerator, and using PI or RI, for semiquantitative analysis of waveforms<sup>21,23-26</sup>.
- The greatest body of scientific evidence has been accumulated for the simple ratio of the MCA-PI divided by the umbilical artery PI (i.e. the CPR), and the second most commonly utilized ratio is its inverse, i.e. the umbilical artery PI divided by the MCA-PI (umbilicocerebral ratio (UCR)).
- When the CPR or UCR are calculated, the measurements from the umbilical artery and MCA should be obtained utilizing the techniques described for these vessels herein. (GOOD PRACTICE POINT)
- The CPR or UCR should be interpreted using gestational-age-related reference ranges<sup>27</sup> rather than a single cut-off. (GOOD PRACTICE POINT)

### What is the appropriate technique for obtaining fetal venous Doppler waveforms?

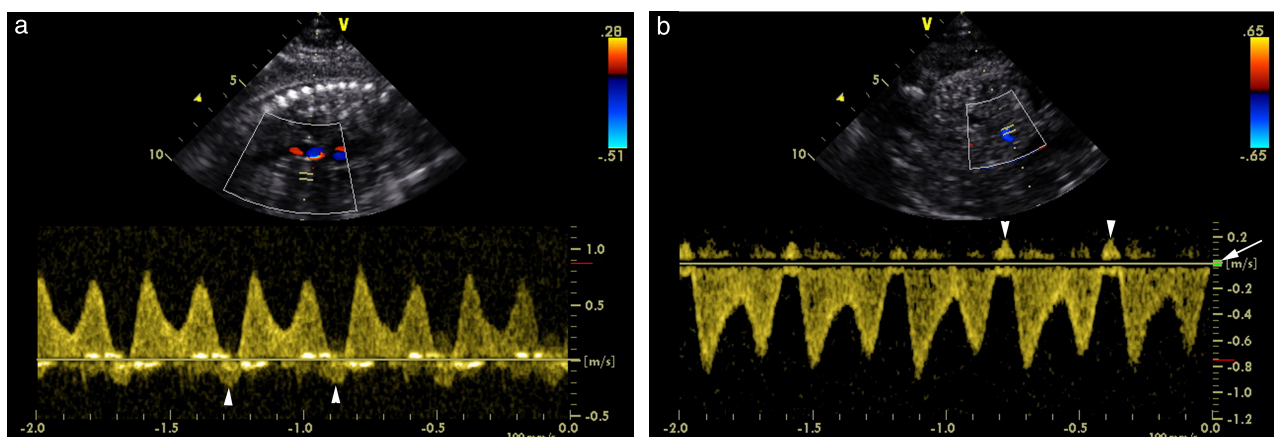
- The ductus venosus (DV) connects the intra-abdominal portion of the umbilical vein to the left portion of the inferior vena cava, just below the diaphragm. The vessel is identified by visualizing this connection by 2D imaging, either in a mid-sagittal longitudinal plane of the fetal trunk or in an oblique transverse plane through the upper abdomen<sup>28</sup>.



- Color flow mapping demonstrating the high velocity at the narrow entrance of the DV confirms its identification and indicates the standard sampling site for Doppler measurements<sup>29</sup>.
- Doppler measurement is best achieved in the sagittal plane from the anterior lower fetal abdomen, since alignment with the ductal isthmus can be well controlled (Figure 8). Sagittal insonation through the chest is also a good option, but more demanding. An oblique section provides reasonable access for an anterior or posterior insonation, yielding robust waveforms but with less control of angle and absolute velocities.
- In early pregnancy and in compromised pregnancies, particular care should be taken to reduce the sample volume appropriately, in order to ensure clean recording of the lowest velocity during atrial contraction (Figure 9).
- The waveform is usually triphasic, but biphasic and non-pulsating recordings, though rarer, may be seen in healthy fetuses<sup>28</sup>.



**Figure 8** Ductus venosus Doppler recording with sagittal insonation aligning with the isthmus portion, without angle correction. The low-velocity vessel wall filter (arrow) does not interfere with the a-wave (a), which is far from the zero line. A high sweep speed allows detailed visualization of variation in velocity.



**Figure 9** (a) Ductus venosus recording showing increased pulsatility at 36 weeks. Interference, including highly echogenic clutter along the zero line, makes it difficult to verify the reversed component during atrial contraction (arrowheads). (b) A repeat recording with slightly increased low-velocity vessel wall filter (arrow) improves quality and allows clear visualization of the reversed velocity component during atrial contraction (arrowheads).

- The velocities are relatively high, between 55 and 90 cm/s, for most of the second half of pregnancy<sup>30</sup>, but are lower in early pregnancy.

#### Which indices should be used?

S/D ratio, RI and PI are the three best known indices to describe arterial flow velocity waveforms. All three are highly correlated. RI and S/D ratio estimate the relationship between PSV and EDV in the Doppler waveform ( $RI = (S - D)/S$ ,  $S/D \text{ ratio} = S/D$ , where S is peak systolic velocity and D is end-diastolic velocity). PI takes into account the PSV, the EDV and the time-averaged mean of the maximum frequency shift over the cardiac cycle ( $PI = (S - D)/TAMX$ , where S is peak systolic velocity, D is end-diastolic velocity and TAMX is the maximum velocity recorded in the MVE averaged over the cardiac cycle; TAMX should not be confused with time-averaged intensity-weighted mean velocity (TAV or  $V_m$ )). In Doppler waveforms showing dynamic changes in the systolic or diastolic components (i.e. in case of uterine artery waveform with presence of notching, or reversed EDV in umbilical artery waveform), PI gives a better estimate of the characteristics of the waveform than do RI or S/D ratio. PI shows a linear correlation with vascular resistance, as opposed to both S/D ratio and RI, which show a parabolic relationship with increasing vascular resistance<sup>31</sup>. Additionally, PI does not approach infinity when there are absent or reversed diastolic values. PI is the index recommended for use in clinical practice and research. (GOOD PRACTICE POINT)

There is currently no high-level evidence to indicate how either CPR or UCR should be utilized in clinical management.

Two indices are described for pulsed-wave Doppler analysis of the veins. The most commonly used is the pulsatility index for veins (PIV)<sup>32</sup>. This is calculated as  $PIV = (V_s - V_a)/TAMX$ , where  $V_s$  is the peak forward velocity during ventricular systole and  $V_a$  is the lowest forward velocity or peak reversed velocity during atrial

contraction (the 'a-wave'). The peak velocity index for veins (PVIV) is reported less frequently and is not featured on most auto-measure packages. PVIV is calculated as  $(V_s - V_a)/V_d$ , where  $V_d$  is the peak forward velocity during atrial contraction (diastole). The use of PIV is recommended in clinical practice. (GOOD PRACTICE POINT)

This Guideline presents the most commonly used techniques in clinical obstetrics, backed by solid scientific documentation. We are aware of important uses and sections of the circulation not mentioned herein, although these vessels and measurements may be of crucial importance in certain individuals. These vessels include, for example, the umbilical vein, hepatic artery, left portal vein and superior vena cava. However, the principles presented in this Guideline are valid for all fetal Doppler examinations.

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#### CITATION

This Guideline should be cited as: 'Bhide A, Acharya G, Baschat A, Bilardo CM, Brezinka C, Cafici D, Ebbing C, Hernandez-Andrade E, Kalache K, Kingdom J, Kiserud T, Kumar S, Lee W, Lees C, Leung KY, Malinger G, Mari G, Prefumo F, Sepulveda W, Trudinger B. ISUOG Practice Guidelines (updated): use of Doppler velocimetry in obstetrics. *Ultrasound Obstet Gynecol* 2021; 58: 331–339.'

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## APPENDIX 1 Levels of evidence and grades of recommendation used in ISUOG Guidelines

### Classification of evidence levels

1++	High-quality meta-analyses, systematic reviews of randomized controlled trials or randomized controlled trials with very low risk of bias
1+	Well-conducted meta-analyses, systematic reviews of randomized controlled trials or randomized controlled trials with low risk of bias
1–	Meta-analyses, systematic reviews of randomized controlled trials or randomized controlled trials with high risk of bias
2++	High-quality systematic reviews of case–control or cohort studies or high-quality case–control or cohort studies with very low risk of confounding, bias or chance and high probability that the relationship is causal
2+	Well-conducted case–control or cohort studies with low risk of confounding, bias or chance and moderate probability that the relationship is causal
2–	Case–control or cohort studies with high risk of confounding, bias or chance and significant risk that the relationship is not causal
3	Non-analytical studies, e.g. case reports, case series
4	Expert opinion

### Grades of recommendation

A	At least one meta-analysis, systematic review or randomized controlled trial rated as 1++ and applicable directly to the target population; or a systematic review of randomized controlled trials or a body of evidence consisting principally of studies rated as 1+ applicable directly to the target population and demonstrating overall consistency of results
B	Body of evidence including studies rated as 2++ applicable directly to the target population and demonstrating overall consistency of results; or evidence extrapolated from studies rated as 1++ or 1+
C	Body of evidence including studies rated as 2+ applicable directly to the target population and demonstrating overall consistency of results; or evidence extrapolated from studies rated as 2++
D	Evidence of level 3 or 4; or evidence extrapolated from studies rated as 2+
Good practice point	Recommended best practice based on the clinical experience of the Guideline Development Group