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Title: STUDY OF THE DEVELOPMENT OF HUMAN PLACENTAL MICROVASCULARITY IN NORMAL GESTATION BY DOPPLER SMI (Superb Microvascular Imaging)

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Keywords: placental vascularization; Doppler SMI (superb microvascular imaging), ultrasound; IUGR (intrauterine growth restriction)

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Abstract: Introduction. To evaluate the development of placental vascularization in normal gestation without pathology by using Doppler superb microvascular imaging (SMI).

Methods. The fetal and maternal parameters of 23 pregnant women without pathology were evaluated at weeks 12, 16, 20-22, 24-26, 28-30, 32-32, 36-38, and 40-42 by 2D ultrasound and color and pulsed Doppler analysis. Doppler SMI was used to evaluate the placental vascularization, pulsatile index (PI) and peak systolic velocity (PV) of the primary (1st), secondary (2nd) and tertiary (3rd) villi, and qualitative placental descriptions and anatomopathological studies of these placentas were performed.

Results. We observed that the number of cotyledons identified by Doppler SMI increased from 2 at 16-18 weeks to an average of 24 between 28 and 38 weeks. We identified the 2nd and 3rd villi starting at 20 weeks of gestation. The PI of the primary villi was constant at approximately 0.8-0.9 in all pregnancies; in contrast, the PI of the secondary and tertiary villi increased from 1.1 to 1.56 and from 1.4 to 1.51, respectively. The PV underwent a significant increase throughout gestation in the secondary and tertiary villi (increasing from 8.9 cm/s to 28.4 cm/s and from 7.5 cm/s to 42.4 cm/s, respectively).

Discussion. We evaluated the development of placental microvascularization using Doppler SMI in pregnancies without pathology and described the normal placental Doppler patterns throughout pregnancy.

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*Conflict of Interest Statement

STUDY OF THE DEVELOPMENT OF HUMAN PLACENTAL MICROVASCULARITY IN NORMAL GESTATION BY DOPPLER SMI (Superb Microvascular Imaging)

Autorship.

Each author must qualify by having participated actively and sufficiently in the study that is being performed and reported.

Conflict of interest.

It doesn't exist any trade association of any author of the text neither any economic benefit with the realization of this work and their publication

Previous publication

The declarations and opinions expressed in the articles and communications belong to the authors and not of the editor or publisher.

IRB approval.

Yes

Permissions

Charts don't exist neither you figure of other authors

Patient consent

I have obtained written patient consent.

The study (0545- N- 18) was approved by the local Ethics and Research Committees on 29-05-2018.

The RCT was registered at ClinicalTrials.gov (NCT03686956).

Fdo Dr Sainz Bueno

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Proyecto Investigación Biomédica

C.P. sainz072018 - C.I. 1001-N-18

20 de julio de 2018

CEIC Hospital Universitario Ntra. Sra. de Valme

RAMON MORILLO VERDUGO
Secretario del CEIC Hospital Universitario Ntra. Sra. de Valme

CERTIFICA

1º. Que el CEIC Hospital Universitario Ntra. Sra. de Valme en su reunión del día 20/07/2018, acta REUNION 7/18 ha evaluado la propuesta del promotor referida al estudio:

Título: EVALUACIÓN DE LA MICROCIRCULACIÓN PLACENTARIA EN LA GESTACIÓN NORMAL Y PATOLÓGICA MEDIANTE ECOGRAFÍA DOPPLER- SMI

Código Promotor: sainz072018 **Código Interno:** 1001-N-18
Promotor: Investigador
Fecha Entrada: 17/07/2018

con el factor de estudio .

1º. Considera que

- El estudio se plantea siguiendo los requisitos de la Ley 14/2007, de 3 de julio, de Investigación Biomédica y su realización es pertinente.
- Se cumplen los requisitos necesarios de idoneidad del protocolo en relación con los objetivos del estudio y están justificados los riesgos y molestias previsibles para el sujeto.
- Son adecuados tanto el procedimiento para obtener el consentimiento informado como la compensación prevista para los sujetos por daños que pudieran derivarse de su participación en el estudio.
- El alcance de las compensaciones económicas previstas no interfiere con el respeto a los postulados éticos.
- La capacidad de los Investigadores y los medios disponibles son apropiados para llevar a cabo el estudio.

2º. Por lo que este CEIC emite un **DICTAMEN FAVORABLE**.

3º. Este CEIC acepta que dicho estudio sea realizado en los siguientes CEIC/Centros por los Investigadores:

CEIC Hospital Universitario Ntra. Sra. de Valme JOSE ANTONIO SAINZ BUENO
(Obstetricia y Ginecología) Hospital Nuestra Señora de Valme

Lo que firmo en Sevilla, a 20 de julio de 2018

Fdo:

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STUDY OF THE DEVELOPMENT OF HUMAN PLACENTAL MICROVASCULARITY IN NORMAL GESTATION BY DOPPLER SMI (Superb Microvascular Imaging)

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Declaration of competing interest
The authors declare no conflicts of interest.

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ABSTRACT.

Introduction. To evaluate the development of placental vascularization in normal gestation without pathology by using Doppler superb microvascular imaging (SMI).

Methods. The fetal and maternal parameters of 23 pregnant women without pathology were evaluated at weeks 12, 16, 20-22, 24-26, 28-30, 32-32, 36-38, and 40-42 by 2D ultrasound and color and pulsed Doppler analysis. Doppler SMI was used to evaluate the placental vascularization, pulsatile index (PI) and peak systolic velocity (PV) of the primary (1st), secondary (2nd) and tertiary (3rd) villi, and qualitative placental descriptions and anatomopathological studies of these placentas were performed.

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Discussion. We evaluated the development of placental microvascularization using Doppler SMI in pregnancies without pathology and described the normal placental Doppler patterns throughout pregnancy.

KEYWORDS: placental vascularization, Doppler SMI (superb microvascular imaging), ultrasound, IUGR (intrauterine growth restriction)

ABBREVIATIONS: SMI (superb microvascular imaging), PI (pulsatile index), PV (peak systolic velocity), IUGR (intrauterine growth restriction), 3D PDA (3D power Doppler angiography), 3D CDI (Color Doppler imaging).

HIGHLIGHTS.

- Through Doppler SMI we were able to identify placental microvascularization.
- From 20-weeks of gestation, we observed increase in villous trees using Doppler SMI
- In primary villi, IP remains constant during pregnancy and the PV increases very little.
- In secondary and tertiary villi, increase in IP and PV is progressive and important

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INTRODUCTION

The placenta is the least understood organ and inescapably one of the most important, not only for sustaining maternal-fetal health during pregnancy but also for the lifelong health of both mother and child. At present, our knowledge about the placenta is quite limited [1]. Placental function and structure affect important aspects of maternal health, such as the development of insulin resistance [2] as well as hypertension during the different stages of pregnancy [3-6]. Placental dysfunction affects the fetus, causing prematurity [7], growth restriction, and neurodevelopmental abnormalities [8].

Alterations in placental implantation and development at the onset of gestation may lead to inadequate placental evolution of trophoblastic invasion and remodeling of the spiral arteries; this developmental alterations may be associated with intrauterine complications that are not clinically evident until the third trimester, such as low birth weight fetuses or preeclampsia [3-8]. Most placental data are obtained from voluntary abortions between 5 and 6 weeks of gestation, leaving knowledge gaps corresponding to the other gestational phases [9].

Despite the described importance of the placenta, research is limited due to the ethical conflicts of obtaining samples of the placenta during its various formative stages. Noninvasive evaluation of the placenta has been attempted without reaching conclusive results. The current clinical tools for detecting placental dysfunction are based on fetal studies, and these lack the sensitivity and specificity to prevent fetal deaths in a cost-effective way [10]. Thus, fetal biometry measures have a sensitivity of 40-74% and a specificity of 72-81% for the diagnosis of a fetus with low weight [11-13]. Umbilical artery (UA) Doppler has a sensitivity and specificity of 19% and 91%, respectively, for predicting low fetal weight in low-risk populations [14] and 55% and 91%, respectively, for high-risk populations [15].

Therefore, most research currently focuses on the study of the placenta in vivo, taking advantage of current techniques to understand placental development, structure, shape and vascularization with good real-time predictive values for fetal adverse effects, to develop preventive measures and treatments that improve the future health of both mother and child. Recently, there has been growing interest in placental studies using 3-dimensional power Doppler (3DPD) And Doppler superb microvascular imaging (SMI). SMI (Canon Medical Systems, Tustin, CA) uses clutter suppression to extract flow signals at rapid frame rates, which provides high-resolution vessel-branching details without the need for contrast agents [16]

Although numerous articles have been published on the application of Doppler SMI for other organs, its diagnostic potential during pregnancy is poorly studied and used. Hasegawa and Suzuki [17] published a study describing the use of SMI to obtain images of a placental infarction. They demonstrated how in a normal placenta, this technology can be used to visualize the vascularization and vascular trees of the cotyledons, while in the placentas of fetuses with severe intrauterine growth restriction (IUGR), the low-velocity placental blood flow cannot be visualized [18].

Therefore, SMI provides new opportunities for the noninvasive characterization of placental vascularization during pregnancy [19]. We intend to evaluate the placental vascularization of normal gestations by Doppler SMI, thus describing normality parameters that will allow its use in pathological situations, such as preeclampsia or IUGR, in the future.

MATERIALS AND METHODS.

We conducted a prospective observational longitudinal study with 26 pregnant women between September 2018 and September 2019 at the Valme University Hospital (Seville, Spain). The study protocol was reviewed and approved by the Ethics Committee of Valme University Hospital (1001-N-18), and informed consent was obtained from all patients.

The 26 pregnant women were all white, underwent spontaneous labor, had single pregnancies without previous obstetric pathology (high blood pressure, diabetes

mellitus, IUGR, adverse neonatal outcomes, morphological and chromosomal abnormalities) or serious maternal comorbidities (autoimmune diseases, heart disease, endocrinopathies, oncological processes, serious infectious processes). They were recruited from the Protocol for Assistance in Pregnancy and Childbirth at Valme University Hospital (Seville, Spain) after verifying that they their combined first-trimester screening indicated that the pregnancy was low risk. Two pregnant women were excluded, one due to the development of gestational diabetes in the second trimester and the other due to the threat of preterm delivery in the third trimester.

The study involved three expert examiners (EA, JC, JS) in fetal ultrasound from the Gynecology and Obstetrics Department of the University Hospital Virgen de Valme. Placental ultrasound captures using Doppler SMI were obtained using a Canon Aplio 500 ultrasound (Tokyo, Japan) with a PUT-675 MV-3D probe. The size of the Doppler sampling window was 35 by 40 mm. The sample size (gate) of the spectral Doppler was 2.5 mm. Captures were made with the patient in the supine decubitus position. Figure 1A shows an example of the images obtained by Doppler SMI.

Fetal and placental parameters were analyzed sequentially at 12, 16, 20-22, 24-26, 28-30, 32-34, 36-38 and 40-42 weeks of gestation. Regarding fetal parameters, biometric data were obtained, fetal weight was estimated by 2D ultrasound and color and pulsed Doppler analysis of the middle cerebral artery (MCA), UA, uterine arteries (UtA) and ductus venosus (DV). The pulsatile index (PI) and the peak systolic velocity (PV) were obtained from all of them.

Regarding the placental parameters, the location of the placenta (anterior or posterior) was determined in all patients except those with any type of anomaly. The placenta was divided into three regions: part 1 (upper part) , part 2 (middle part) and part 3 (lower part). Figure 1B shows an outline of the placental division performed.
For all placental regions, placental morphological characteristics were analyzed: the number of cotyledons, number of primary villous trunks and number of secondary and tertiary villous trunks. The following placental parameters were evaluated by Doppler SMI: the amount of secondary and tertiary villi was classified as abundant or sparse (abundant if they occupied more than 50% of the Doppler gate). Subsequently, the blood flow (PV and PI) of the primary, secondary and tertiary villous trunks of the three

parts of the placenta was analyzed. The flows of the maternal placental basal plate and the fetal placental chorionic plate were also analyzed.

Anatomopathological placenta study were evaluated based on the latest international criteria for placental lesions as described in the Amsterdam Placental Workshop Group Consensus Statement [20]. The placentas were weighted; percentile was determined by specific placenta weight tables [21]. For each placenta, at least five full-thickness disk samples were taken. Additional samples were obtained from the membrane roll, the umbilical cord, and any other abnormal area. Samples were embedded in paraffin blocks. Five-micrometer-thick slices were than H&E stained for microscopic assessment. A histopathological analysis was performed to demonstrate whether there was data of maternal vascular malperfusion and fetal vascular malperfusion.

RESULTS

Twenty-four pregnant women were evaluated (2 were excluded due to gestational diabetes and threat of premature labor, respectively). Regarding the epidemiological data, 79.1% (19) of the pregnant women were nulliparous with an average age of 30.5+/− 4.1 years, 16.6% (4) were smokers, and the mean BMI was 24.0+/− 4.1. The pregnancies presented mean BHCG and PAPPA values in the first trimester of 1.4+/− 0.8 and 1.2+/− 0.5 MoM, respectively. We evaluated 12 anterior (50%) and 12 posterior (50%) placentas. Of the births, 83.3% (20) were spontaneous, 16.25% (4) were instrumented deliveries, and 4.1% (1) were cesarean deliveries. There were a total of 17 (70.8%) males among the neonates, and the average birth weight was 3240+/− 407 grams. A total of 79.1% (19) of the newborns had a UA pH at birth > 7.2, and there was one case of admission to neonatology due to respiratory distress.

Table 1A shows the results of the subjective quantitative evaluation of placental parameters evaluated by Doppler SMI. We observed that the number of cotyledons identified by Doppler SMI throughout pregnancy increased from 1 cotyledon identified at week 12 to 3 cotyledons at week 16, 18 cotyledons at week 20 (6 for each evaluated placental area), 24 (8 for each placental area evaluated) in weeks 28, 32, and 36, and

finally, in week 40, there were 21 (7 for each evaluated placental area). Secondary and tertiary villi were identifiable after 20 weeks (classified as abundant in 80% and 3.8% of the placentas, respectively, at this gestational time point), and there was a clear increase between 28 and 38 weeks, especially in the tertiary villi (considered abundant in 23.1% of the placentas). Both secondary and tertiary villi (classified as abundant in 30.8% and 3.8% of placentas, respectively) slightly decreased at the end of gestation (40-42 weeks).

The results obtained from the objective quantitative placental assessment by Doppler SMI are presented in Table 1B by gestational weeks and the different parts of the placenta. We observed that the PI at the basal plate remained constant (between 0.5-0.6) between 12 and 38 weeks of gestation and increased between 40 and 42 weeks of gestation (1.1). In the chorionic plate, the PI increased progressively from 0.75 at 12 weeks of gestation to 0.92 at 40 weeks of gestation. The identification of the Doppler flow in the primary villi could be performed starting at 12 weeks of gestation; in contrast, the Doppler flows in the secondary and tertiary villi were identifiable after 20 weeks of gestation. The PI of the primary villi remained constant at approximately 0.8-0.9 throughout pregnancy, while the PI of the secondary and tertiary villi increased from 1.1 to 1.56 and from 1.4 to 1.51, respectively. Between weeks 40 and 42, the tertiary villi underwent a significant increase in PI to 3.79.

The peak velocity in the basal plate decreased slightly throughout pregnancy (from 31.1 cm/s at 12 weeks of gestation to 22.4 cm/s at 40 weeks of gestation), while in the chorionic plate, the peak velocity increased from 11.7 cm/s at 12 weeks of gestation to 14.5 cm/s at 40 weeks of gestation. The peak velocity in the chorionic villi increased slightly in the primary villi (from 11.2 at 12 weeks of gestation to 14.4 cm/s at 40 weeks) and significantly in the secondary and tertiary villi (from 8.9 cm/s to 28.4 cm/s in the secondary villi and from 7.5 cm/s to 42.4 cm/s in the tertiary villi at 20 and 40 weeks gestation, respectively,).

Figures 2 and 3 show the evolution of the PI and PV throughout pregnancy, evaluated by Doppler SMI in the chorionic plate, basal plate, primary villus, secondary villus and tertiary villus. Figure 4 shows the qualitative evaluation of placental branching as captured with the Doppler SMI.

Maternal and fetal Doppler yielded the following results throughout pregnancy: the mean PI of the UtA was 1.03+/– 0.3 at 20 weeks, changing to 0.77+/– 0.2 at 32 weeks and 0.69+/– 0.1 at 40 weeks. The mean PI of the UA was 1.22+/– 0.2 at 20 weeks, increasing to 0.83+/– 0.1 at 32 weeks and 0.71+/– 0.1 at 40 weeks. The mean PI of the UA was 1.22+/– 0.2 at 20 weeks, increasing to 0.83+/– 0.1 at 32 weeks and 0.71+/– 0.1 at 40 weeks. The MCA and venous ductus presented PI values of 1.66+/– 0.4 and 0.75+/– 0.2 at 20 weeks, 1.82+/– 0.4 and 0.58+/– 0.2 at 32 weeks and 1.26+/– 0.2 and 0.60+/– 0.1 at 40 weeks, respectively.

Regarding the anatomopathological data of the postdelivery placentas, twenty-one placentas were evaluated. Table 2 shows the data of macroscopic placental study, maternal vascular malperfusion of the placental bed and fetal vascular malperfusion.

DISCUSSION

The main finding of our study was that it is possible to assess the development of placental microvascularization throughout pregnancy using Doppler SMI, thus allowing us to describe the normal placental parameters in pathology-free pregnancies. We were able to qualitatively and quantitatively assess the degree of placental vascular branching at the different gestational time points under study.

Real-time measurements of placental flow have previously been attempted using power Doppler and 3D power Doppler angiography (3D PDA) [22-24]. Welsh et al. [22] designed software using power Doppler measurements of a cotyledon with normal vascularization and a cotyledon isolated from the circulation by a blood clot and found lower flow measurements in the isolated cotyledon. Konje et al. described a prototype using placental images of different gestations taken with 3D PDA [23]. Rizzo et al. [24] quantified placental vascularization between 11 and 14 gestational weeks by 3D PDA, observing that pregnancies with abnormal karyotypes had less volume and placental vascularization. In his study, Campbell presented placental capture using Color Doppler imaging (3D CDI) and 3D PDA. He demonstrated that the flow velocity in the primary

villi was lower, while the flow volume in the tertiary villi in a normal pregnancy was surprisingly high; this finding indicates the promising potential to quantify these parameters since this technique could identify placentas with vascular damage leading to fetuses with IUGR [25].

Previous articles have attempted to relate the measurement of placental vascular indices with pathology such as preeclampsia. Eastwood et al. [26], in their measurements by 3D PDA, found a decreased vascularization and flow index in the first trimester in patients who developed preeclampsia compared to those with normal pregnancies. In Plasencia et al., the results of the vascular indices were similar [27].

However, there are critics of these ultrasound techniques. The Raine-Fenning group has published several articles related to 3D power Doppler and its application to the measurement of placental vascularization [28-30]. They concluded that the measurement of placental flow by 3D power Doppler has low intra- and interobserver reproducibility, stating that the technique has a limited role in both clinical application and research. The lack of reproducibility reduces the technique's ability to differentiate a pathological placenta from a normal placenta since its results can be highly variable due simply to errors in the acquisition and processing of the data. Therefore, it is unlikely that 3D power Doppler will have clinical value in the second and third trimesters of pregnancy [28].

In the literature, we found articles that describe the different types of maternal-fetal hypoxia: preplacental hypoxia, uteroplacental hypoxia, and postplacental hypoxia [31]. In uteroplacental hypoxia, maternal oxygenation is normal, but due to damage to the uteroplacental circulation, both the fetus and the placenta are hypoxic [32]. In this situation, the peripheral placental villi have a highly developed network of peripheral capillaries, and the fetal blood flow is normal or reduced. In postplacental hypoxia, the fetus is hypoxic, while the mother is not, and the placenta may even have elevated oxygen levels, a phenomenon described as a hyperoxic placenta. In this situation, the villous capillaries are poorly developed, and placental branching is absent. Ultrafast Doppler technology discriminates the placental from the and maternal circulation, detecting and differentiating these two hemodynamic situations. Clinical studies with ultrafast Doppler ultrasound could confirm the hypothesis of uteroplacental and

postplacental hypoxia and help to understand why preeclampsia and restricted intrauterine growth are associated [33].

The qualitative analysis of our study shows how the degree of branching and the placental vascular network increase as pregnancy progresses (Figure 4). Between weeks 12 and 20, we were able to identify only primary villous trunks, and we could not visualize their branching. At week 20, however, we were able to identify both primary villous trunks and their branching into secondary and tertiary villus trunks.
While the secondary villi are abundant from week 20 of gestation and remain stable throughout pregnancy, the tertiary villi reach their maximum development between weeks 28 and 36 and could be visualized in our captures with Doppler SMI. We believe that the lower percentage of placentas with an abundance of this type of villi at week 40 is due to the difficulty of measurement and not to the fact that the vascular network actually decreases (Table 1).

There are some publications that, like our study, define the placental microcirculation pattern and vascular capillary density. Reynolds et al. described in a sheep model that as the microcirculation pattern of the maternal placental side develops, the diameter of the capillaries increases as pregnancy progresses, while the capillary density is modest, on the order of 1.5 to 3 times the density at the beginning of pregnancy. In contrast, in the fetal surface of the placenta, the diameter of the capillaries decreases as the pregnancy progresses, while the capillary density increases on the order of 6 to 12 times the initial density [34]. Using a sheep model of pathological pregnancies, Reynolds and his group, using their own results and those of Mayhew et al. [35], showed that angiogenesis was altered, resulting in a reduction of placental vascular development that was associated with reduced flow in the fetal and maternal sides of the placenta.

Regarding the placental quantitative analysis, the number of cotyledons (the functional unit) increases as pregnancy progresses, reaching its maximum at week 32.
When analyzing the fetal chorionic plate and the maternal basal plate, we observed that the PI in the maternal placental side maintains lower values compared to the PI of the fetal placental side throughout the entire pregnancy, while in contrast, the PV is higher in the maternal plate than in the fetal plate (Figure 2).

With respect to the quantitative objective data (Table 1), we showed that the primary and secondary villi maintain a close developmental trend regarding their vascular indices, while the tertiary villi follow a different pattern. The PI decreases in the primary and secondary villi throughout most of pregnancy and then slightly increases at the end of pregnancy, while the tertiary villi follows a different pattern, maintaining higher values than the primary and secondary villi throughout the entire pregnancy (Figures 2 and 3).
PV increases progressively throughout pregnancy in the three parts of the placenta and in the three vascular trunks of placental branching, with lower velocities observed in larger-diameter vessels and primary villi and higher velocities observed in the tertiary villi (Figures 2 and 3).

Like our group, LeGallo described the cotyledon as a functional and anatomical unit of fetal circulation. Each cotyledon consists of a main villous trunk and its branches. Two types of angiogenesis are described. Angiogenesis with branching occurs until approximately week 24 and is responsible for the development of the placental flow system. In the third trimester, angiogenesis without branching predominates and is correlated with the formation of mature intermediate villi that increase in length [36].

In our study, we observed this growth pattern because, as we have previously noted, we observed an increase in the number of cotyledons in weeks 20-28 that peaked at week 32, and we observed the appearance branching of the villous trunks. In the ultrasound performed in the third trimester, we observed a pause in the increase in the number of cotyledons, although villous density increased and reached its maximum during weeks 28 to 36.

Our next objective is to extrapolate these normality parameters to pathological pregnancies in order to compare them and determine whether we are truly capable of identifying early differences in placental indices and performing a qualitative assessment of the placenta. As we have previously mentioned, the introduction of new technologies such as ultrafast Doppler [33,37] and Doppler SMI, which is what we used in the present study, allows the evaluation of placental vasculature without image artifacts, thus providing a new noninvasive tool for assessing placental functioning.

We found limitations in our study, such as the difficulty of obtaining measurements during the last weeks of gestation due to fetal size, which hinders access to the placenta. Likewise, we found greater difficulty in the measurement of the posterior placenta, which was less accessible to us.

Ethics statement

Written patient consent was obtained for publication of this research. The research was granted approval by the Ethics Committee of Valme University Hospital (1001-N-18)

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Figure Legend

Table 1. 1a: Results of the subjective quantitative evaluation of placental parameters evaluated by Doppler SMI. 1b: Results of the objective quantitative evaluation of placental parameters evaluated by Doppler SMI.

Table 2: Placental macroscopic description, maternal vascular malperfusion of the placental bed and fetal vascular malperfusion.

Figure 1 Fig 1a (left): Placental capture of 20 week pregnancy where we can identify the placental arborization. Fig 1b (right): The arrangement of the placenta (red) in the relation to the maternal cranio-caudal axis is shown in the outline. The next image shows placental división, and the last one shows a functional unit.

Figure 2. Pulsatility index (PI) evolution throughout pregnancy in the different parts. A) chorionic and basal plate, B) primary villus, C) secondary villus and D) tertiary villus

Figure 3. Peak velocity (PV) evolution throughout pregnancy in the different parts. A) chorionic and basal plate, B) primary villus, C) secondary villus and D) tertiary villus

Figure 4. Image captures of placental arborization evaluated by Doppler SMI throughout pregnancy.

Table

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Table 1. 1a: Results of the subjective quantitative evaluation of placental parameters evaluated by Doppler SMI. 1b: Results of the objective quantitative evaluation of placental parameters evaluated by Doppler SMI.

Table1a		WEEK 12-14 N:24	WEEK 16-18 N:24	WEEK 20-22 N:24	WEEK 24-26 N:22	WEEK 28-30 N:23	WEEK 32-34 N:22	WEEK 36-38 N:22	WEEK 40-42 N:22
		Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage
No of cotyledons/ No of primary villi									
	Part 1 (upper part)	-	-	4.96±1.50	6.36±1.60	7.57±2.08	7.82±2.42	7.68±2.60	6.50±2.17
	Part 2 (middle part)	1.00±1.10	2.71±0.75	6.27±2.08	6.77±2.22	7.30±1.74	7.95±2.51	8.40±2.81	6.33±2.06
	Part 3 (lower part)	-	-	4.77±1.39	6.23±2.24	6.70±2.32	6.82±2.85	6.95±1.98	6.11±2.02
No of villi *									
Secondary	**Part 1 (upper part)**	-	-	20(80.8%)	20(76.9%)	21(80.8%)	20(76.9%)	19(73.1%)	8(30.8%)
	Part 2 (middle part)	-	-	20(80.8%)	21(80.8%)	21(80.8%)	20(76.9%)	18(69.2%)	7(26.9%)
	Part 3 (lower part)	-	-	19(76.9%)	20(76.9%)	21(80.8%)	19(73.1%)	18(69.2%)	6(23.1%)
Tertiary	**Part 1 (upper part)**	-	-	1(3.8%)	3(11.5%)	5(19.2%)	6(23.1%)	7(26.9%)	1(3.8%)
	Part 2 (middle part)	-	-	1(3.8%)	2(7.7%)	5(19.2%)	6(23.1%)	7(26.9%)	1(3.8%)
	Part 3 (lower part)	-	-	1(3.8%)	3(11.5%)	5(19.2%)	6(23.1%)	7(26.9%)	1(3.8%)

Table 1b.		WEEK 12-14 N:24	WEEK 16-18 N:24	WEEK 20-22 N:24	WEEK 24-26 N:22	WEEK 28-30 N:23	WEEK 32-34 N:22	WEEK 36-38 N:22	WEEK 40-42 N:22
		Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage	Mean / Percentage
Pulsatile index									
	Basal plate (maternal)	0.63±0.12	0.50±0.20	0.50±0.33	0.52±0.43	0.54±0.52	0.58±0.61	0.67±0.76	1.11±2.28
	Chorionic plate (fetal)	0.75±0.20	1.29±0.60	1.23±0.84	1.10±0.42	0.83±0.76	1.06±0.76	0.96±0.89	0.92±0.46
Primary villi									
	Part 1 (upper part)	-	-	1.07±1.05	0.73±0.36	1.31±2.49	0.80±0.55	1.01±0.75	0.66±0.34
	Part 2 (middle part)	0.89±0.70	1.33±0.77	1.09±1.37	0.86±0.85	0.88±0.44	0.77±0.56	0.80±0.55	1.03±0.82
	Part 3 (lower part)	-	-	1.20±1.46	1.07±1.00	1.14±0.74	1.04±0.67	1.21±1.52	1.65±1.77
Secondary villi									
	Part 1 (upper part)	-	-	0.99±0.57	0.90±0.82	1.43±1.73	1.56±2.45	0.92±0.74	1.52±1.61
	Part 2 (middle part)	-	-	1.12±1.99	1.44±1.66	1.10±0.82	1.23±2.39	1.35±1.70	1.56±1.19
	Part 3 (lower part)	-	-	1.19±1.94	0.96±0.96	1.04±1.03	1.47±1.14	1.11±1.17	1.46±1.52
Tertiary villi									
	Part 1 (upper part)	-	-	1.43±1.08	1.25±1.27	1.79±1.54	2.41±4.40	1.34±1.92	3.79±3.32
	Part 2 (middle part)	-	-	1.41±1.48	1.59±2.01	1.81±2.16	1.34±1.56	1.51±1.53	7.32±7.32
	Part 3 (lower part)	-	-	1.42±1.10	1.68±1.38	1.27±1.22	1.42±1.61	3.14±4.73	1.71±1.59
Peak systolic velocity +									
	Basal plate (maternal)	31.17±18.84	37.01±10.21	23.95±12.57	20.43±9.14	30.27±23.91	21.07±12.57	20.62±12.35	22.41±16.15
	Chorionic plate (fetal)	11.76±3.48	9.70±2.05	11.87±3.69	13.67±2.69	15.80±5.05	20.23±27.68	14.73±5.60	14.59±8.18
Primary villi									
	Part 1 (upper part)	-	-	10.07±2.83	14.31±8.15	15.73±6.45	14.39±5.18	13.78±5.01	14.81±2.55
	Part 2 (middle part)	11.23±3.50	10.84±2.31	10.17±4.45	12.59±4.07	15.21±7.64	13.46±3.60	14.48±4.71	11.40±2.66
	Part 3 (lower part)	-	-	9.33±2.95	13.06±3.04	14.24±8.84	14.06±4.88	14.52±5.07	22.67±21.41
Secondary villi									
	Part 1 (upper part)	-	-	9.23±3.34	9.14±2.57	13.19±6.50	9.88±3.20	11.79±4.49	13.55±7.14
	Part 2 (middle part)	-	-	8.95±3.68	10.48±5.11	10.35±2.93	9.46±3.89	11.77±4.25	28.48±31.86
	Part 3 (lower part)	-	-	8.60±3.28	9.71±3.40	11.48±4.53	11.08±3.09	11.37±3.13	11.03±1.90
Tertiary villi									

	Part 1 (upper part)	-	-	8.03±2.78	8.46±4.06	8.00±2.35	8.31±2.93	12.66±11.00	22.92±30.51
	Part 2 (middle part)	-	-	7.52±2.75	8.43±3.93	9.03±5.50	9.18±4.10	13.51±15.17	42.48±64.76
	Part 3 (lower part)	-	-	7.66±2.47	7.77±2.85	9.16±4.00	8.72±3.13	8.64±2.91	9.92±5.15

Note: * Expressed as a percentage of placentas with abundant development of secondary and tertiary villi.
+ Expressed in cm/seg peak systolic velocity value

Table 2: Placental macroscopic description, maternal vascular malperfusion of the placental bed and fetal vascular malperfusion.

Placental classification	Histopathological feature	Frecuency Mean / Percentage
Macroscopic description	Gestational week	40 ± 0.94
	Placental weight (g).	613.5 ± 119
	Placental weight < 10^{Th} percentile for gestational age.	0 (0%)
	Macroscopic lesions	4 (19%)
Maternal stromal-vascular lesions-malperfusion	Global/partial—late: accelerated villous maturation	0 (0%)
	Microinfarct/segmental/complete—villous infarct	11 (52%)
	Retroplacental hemorrhage	0 (0%)
	Decidual arteriopathy (mural hypertrophy)	8 (38%)
Fetal stromal-vascular lesions—malperfusion	Segmental/complete—chorionic plate or stem villous thrombi	2 (9.5%)
	Avascular villi	0 (0%)
	Villous stromal-vascular karyorrhexis	2 (9.5%)
	Complete/ partial stem vessel obliteration	5 (23.8%)

Figure

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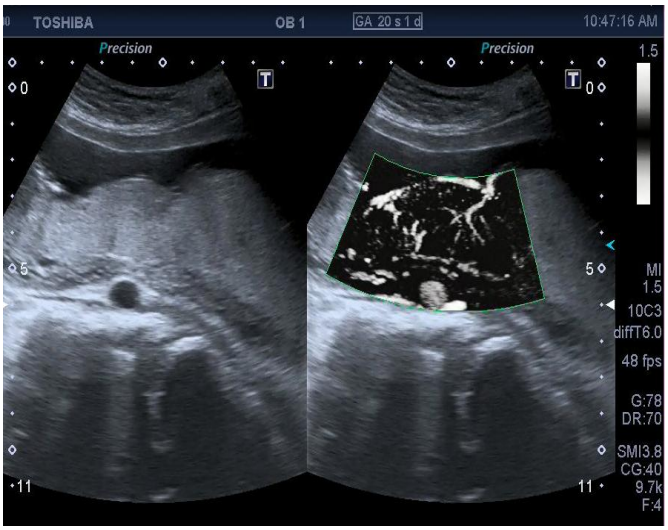

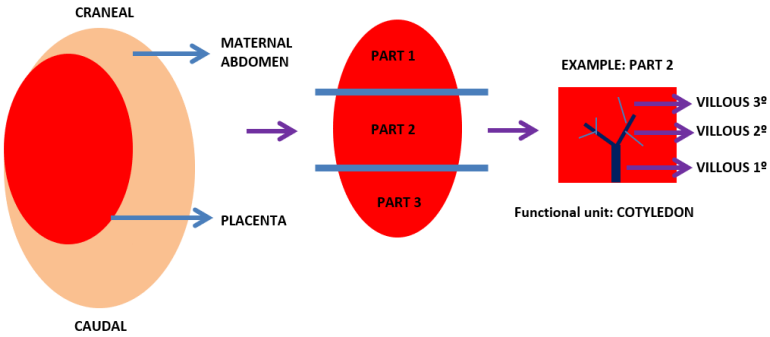

Figure 1

Figure
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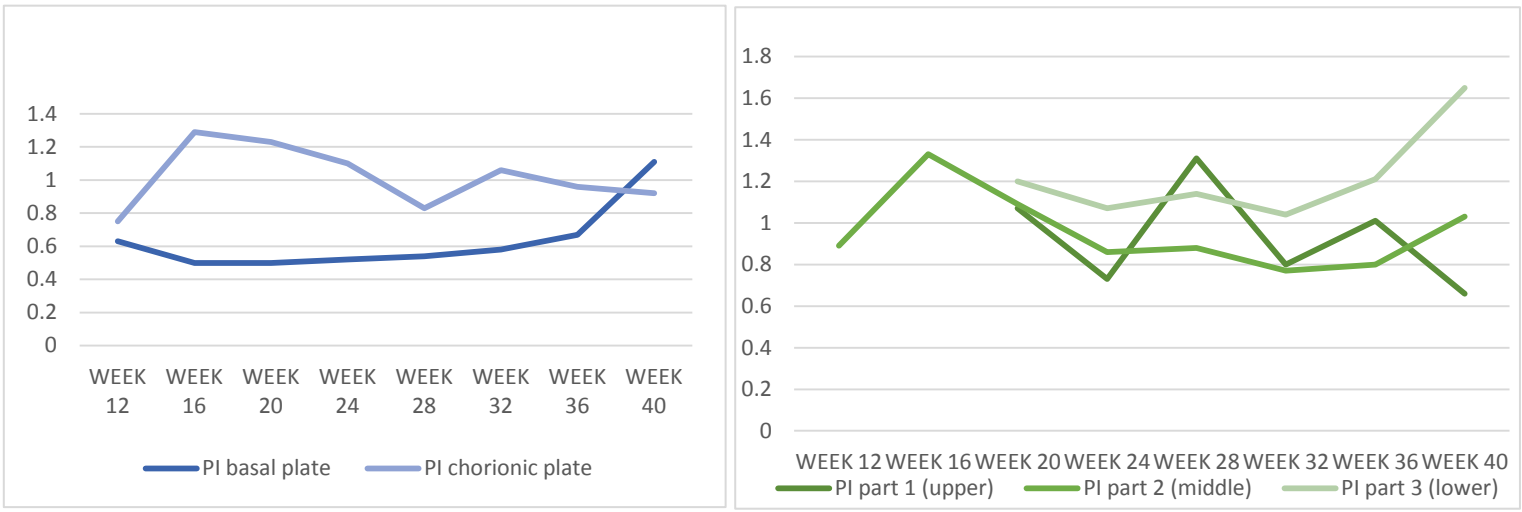

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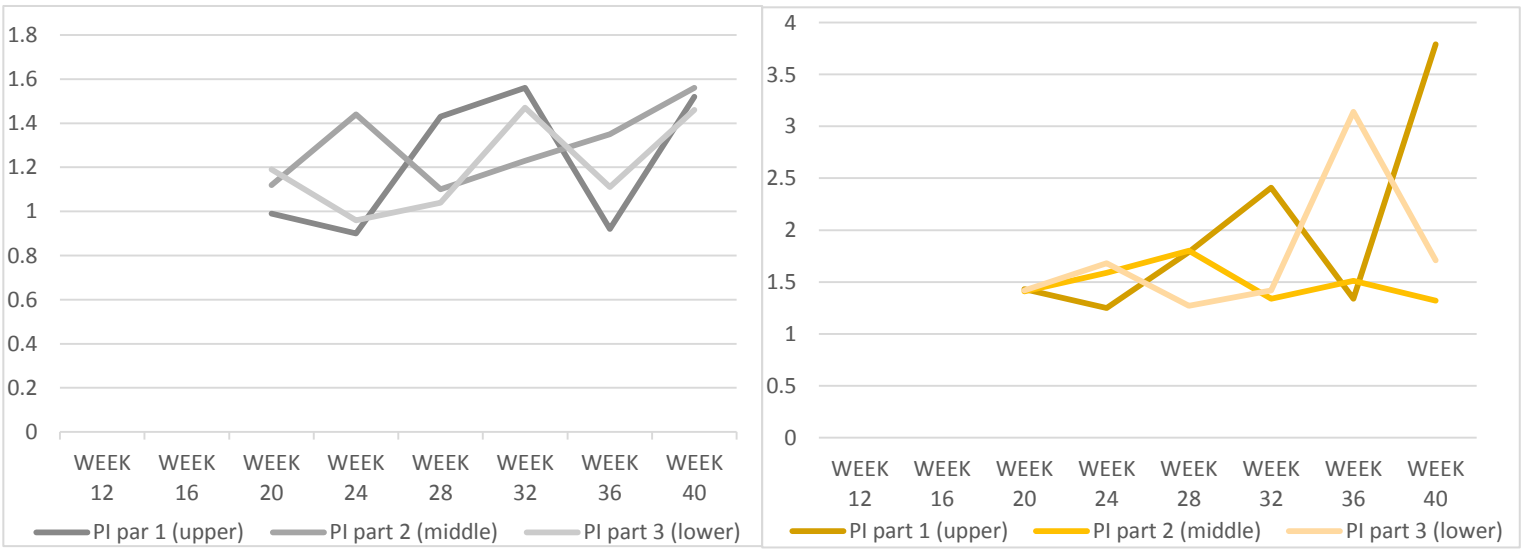

Figure

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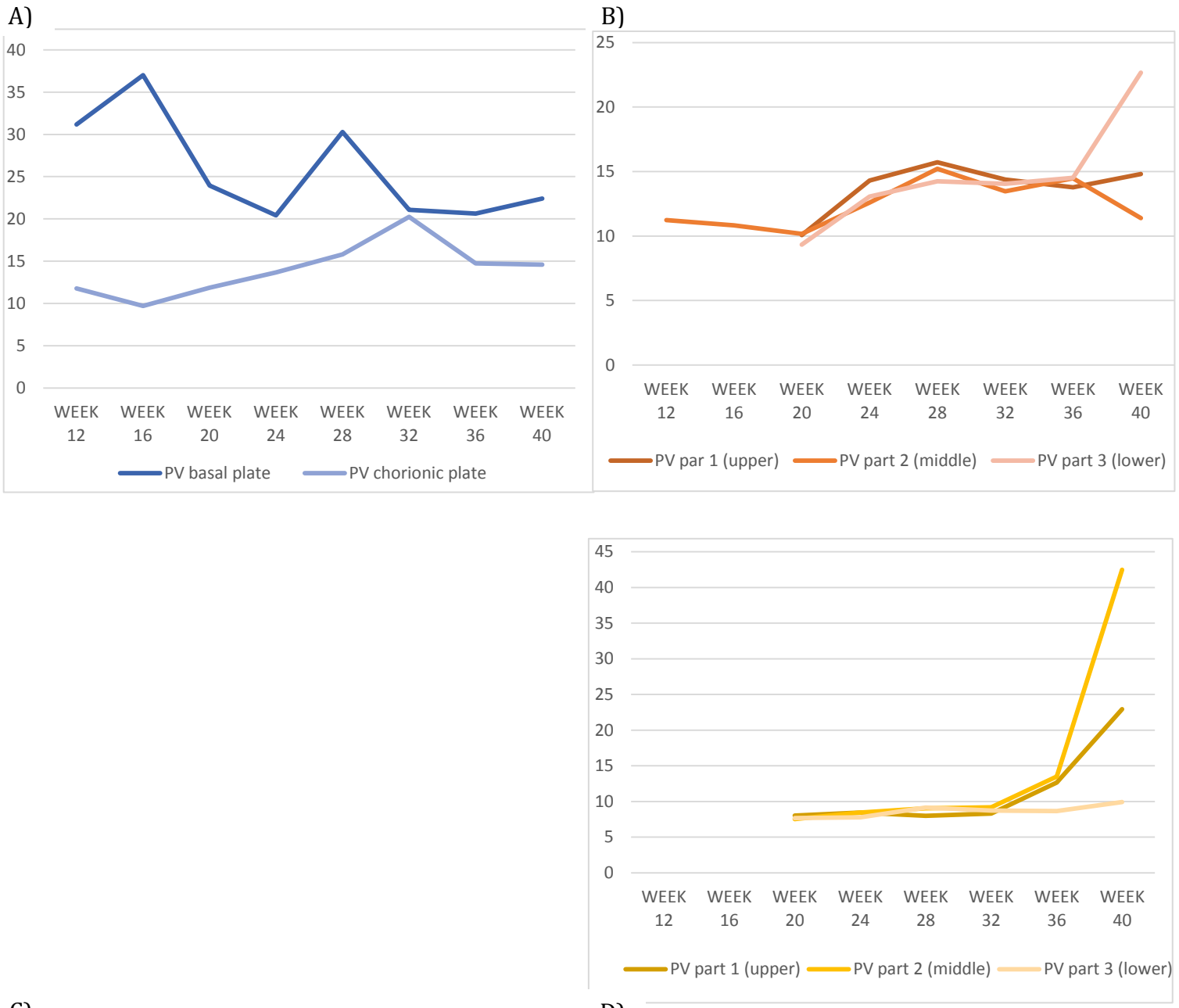

Figure 3.

Figure

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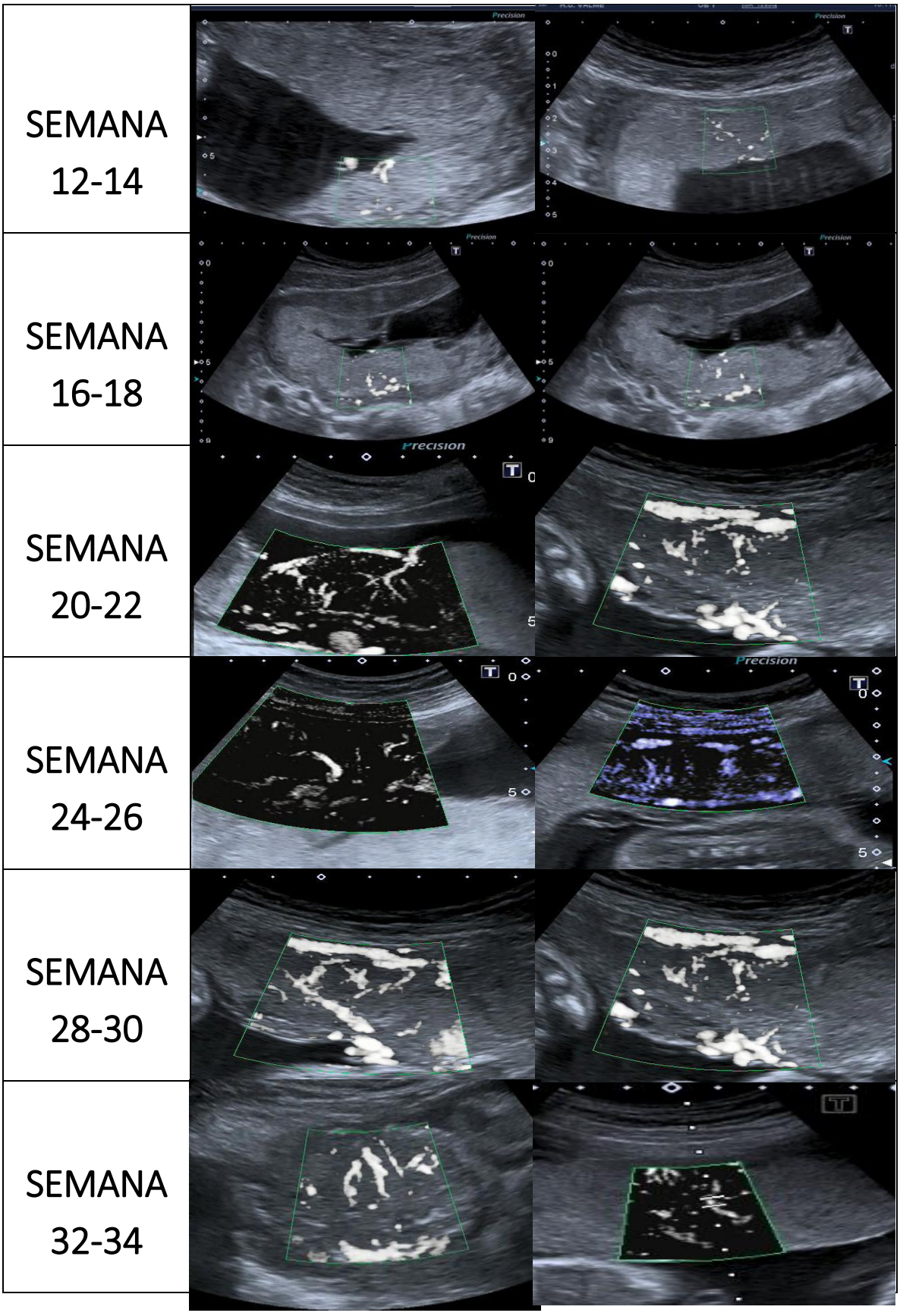
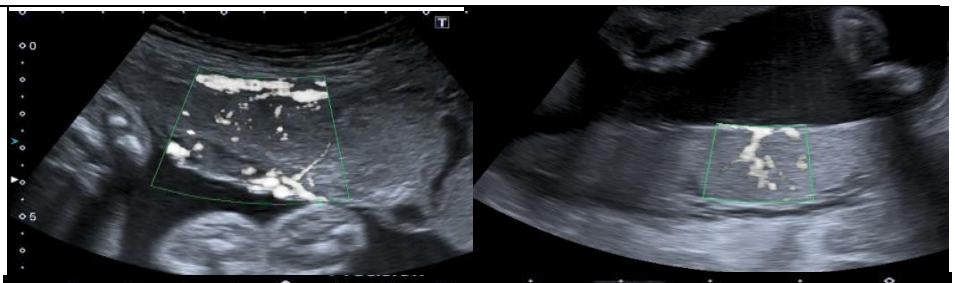
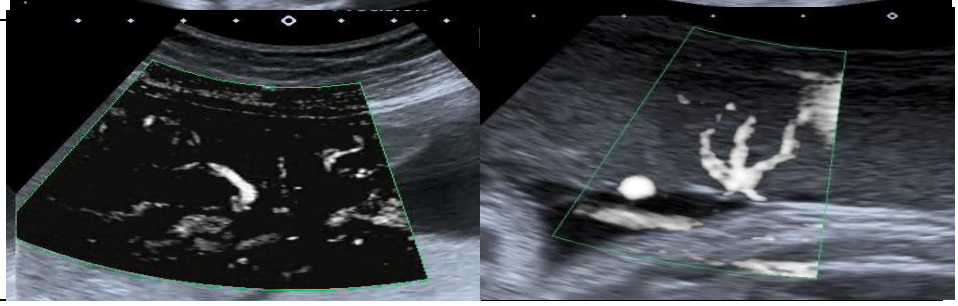


SEMANA 36-38	
SEMANA 40-42	

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