Maternal influence on fetal size and use of longitudinal fetal surveillance in predicting adverse perinatal outcomes

Henriette Odland Karlsen

Dissertation for the degree of philosophiae doctor (PhD)
at the University of Bergen

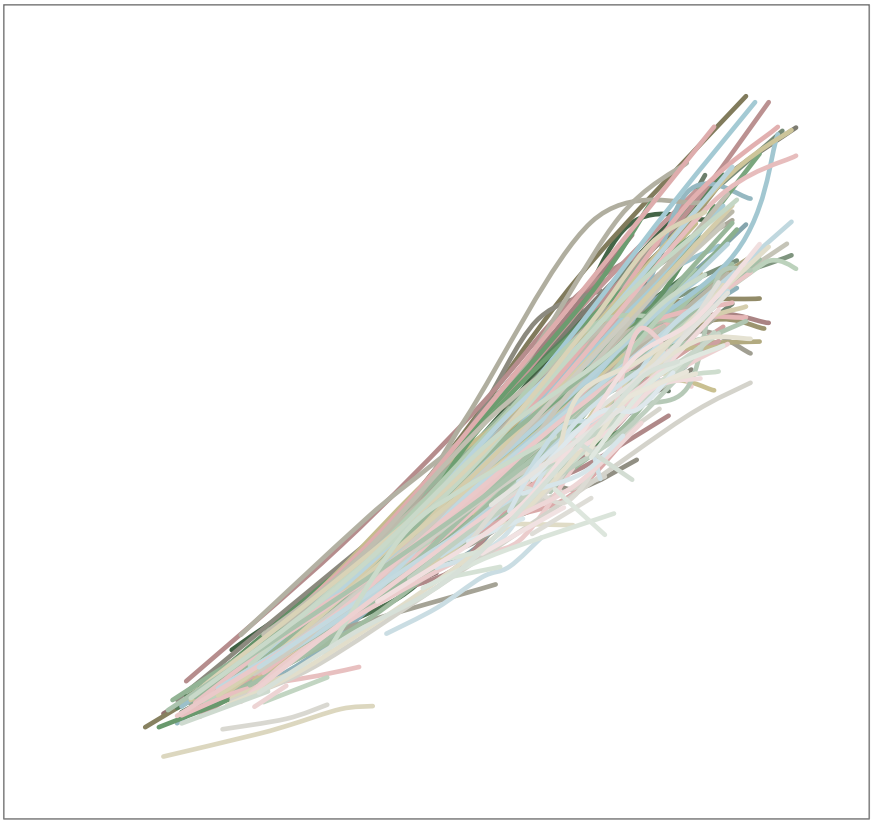
2016

Dissertation date: 15.06.2016

Scientific environment

Clinical Fetal Physiology Research Group, Department of clinical science, University of Bergen

Fetal Medicine Unit, Department of Obstetrics and Gynecology, Haukeland University Hospital, Bergen



Contents

Page

Acknowledgements

The present work was carried out at the Section for Fetal Medicine and Ultrasound Diagnostics, Department of Obstetrics and Gynecology, Haukeland University Hospital and at the Department of Clinical Medicine, University of Bergen. I offer my sincere thanks to the heads of the department Per Børdahl and Susanne Albrechtsen and section leader Synnøve Lian Johnsen who provided the facilities and support necessary to accomplish the project. From January 2012 I have been founded by a scholarship from the University of Bergen. Several people have contributed to this work and I would like to thank all of you.

Synnøve Lian Johnsen, my co-supervisor from the very first beginning until you became my principal supervisor from June 2014. I am very grateful for your contagiously encouragement and I always feel very motivated after our meetings. I appreciated that you enabled me to combine research and clinical work, and you have been a great support in both settings. Thank you for let me be a part of The Fetal Medicine Unit and for believing in me.

Torvid Kiserud, my principal supervisor until June 2014, and further as co-supervisor to the very end. Thank you for inviting me into your inspiring research group. I appreciate your wide experience and your ability to think in new ways. Your linguistic talent is enviable. Despite the fact that you are a very busy man, I have always felt that I have been given priority and your thorough and quick revisions have been very valuable.

Svein Rasmussen, my co-supervisor and my statistical advisor. Thank you for patiently trying to learn me somewhat complicated statistics. Your contribution into this has been of great value.

I also want to express my thanks to Cathrine Ebbing for your contribution in paper III. I appreciates that your door is always open and for letting me bother you with all my professional and private concerns, your support has been very valuable.

My friend and colleague Liv Cecilie Thomsen, you have contributed to this thesis in several ways by shearing of your own research experience and your knowledge on how to write a thesis. I also appreciate your care and our meetings outside Kvinneklinkken.

Per Børdahl, thank you for your support in the very beginning of my research carrier. I appreciate your engagement when I asked for help finding literature of obstetric history.

I am very grateful to have been allowed to work with so many good colleagues at The Fetal Medicine Unit, all have been very helpful and supportive. I will thanks the midwifes for their eagerly help with recruitment. Jorunn, Anita and the office staff deserves thanks for making the days go smoothly. Especially thanks to all the doctors at the Fetal Medicine Unit for patiently giving me professional advice and for shearing of their experience.

Thanks to all the women who agreed to participation and made this study possible.

To all of my valuable friends, thank you for all your support over the last years. Whiteout you my life had been considerably less fun.

Finally, my warmest tanks to my always supporting parents, I am grateful for all your help and for believing in me. My dearest two children, Magnus and Martin, you are my bright spots in life, and I am thankful for your patience and love. Finally I also want to thank Frank; you have managed to distract me from tough days and contributed to a lot of joy. Thank you for all your patience and support.

List of abbreviations

AC	Abdominal circumference
AED	Absent end diastole
AGA	Appropriate for gestational age
ARED	Absent and reversed end diastole
aRR	Adjusted relative risk
BMI	Body mass index
BPD	Biparietal diameter
CI	Confidence interval
CPR	Cerebroplacental ratio
CRL	Crown rump length
CTG	Cardiotocography
DV	Ductus venosus
EFW	Estimated fetal weight
FL	Femur length
HC	Head circumference
IgF	Insulin-like growth factor
IUGR	Intrauterine growth restriction
IVF	In vitro fertilization
LGA	Large for gestational age
LMP	Last menstrual period
MCA	Middle cerebral artery
MBR	Medical birth registry
NICU	Neonatal intensive care unit
NPV	Negative predictive value
PE	Pre-eclampsia
PI	Pulsatility index
PPV	Positive predictive value
RR	Relative risk
SD	Standard deviation

SGA	Small for gestational age
SF	Symphysis fundal
SPSS	Statistical package for the social sciences
UA	Umbilical artery
UtA	Uterine artery
VIF	Variance inflation factor
WHO	World Health Organization

Abstract

Background: Fetal growth is influenced by maternal factors, but also more specifically determined by level of fetoplacental circulation. We tested the hypothesis that fetal abdominal circumference (AC) in the second trimester was influenced by maternal weight gain during pregnancy. Further, we investigated whether fetal growth assessment using serial measurements, i.e. conditional centiles for estimated fetal weight (EFW), improved the prediction of perinatal outcomes in a population at high risk of having small for gestational age (SGA) fetuses. Similarly, we tested the use of conditional centiles for the middle cerebral artery (MCA) pulsatility index (PI) and the cerebroplacental ratio (CPR).

Material and method: In study I data from the «Fetal age and growth» study that included healthy pregnant women, were used to examine the effect of maternal weight gain on fetal AC at gestational week 15-25. Z-scores were used in a linear regression analysis. In study II and III pregnant women at risk of, or diagnosed with an SGA ($\leq 5^{th}$ centile) fetus were included for serial ultrasound measurements of fetal size and Doppler. In both studies data from the final two examinations were included in a regression analysis. Adverse outcomes were birth <37 weeks, operative delivery due to fetal distress, 5-min Apgar score <7, neonatal hypoglycemia (glucose <2.0 mmol/L), admission to the neonatal intensive care unit, and perinatal mortality.

Results: Study I: Complete data were available in 515 of the 650 included women. We found a positive association between z-score for weekly maternal weight gain and z-score for second trimester fetal AC ($p = 0.001$). Study II and III: Complete biometric data were obtained for 211 women and serial Doppler measurements were available in 207 participants. Combining conditional and conventional centiles of EFW ≤5 and ≤10 (Study II) and CPR ≤10 (Study III) significantly improved the prediction of adverse outcomes compared with conventional centiles for EFW and CPR alone.

Conclusions: Study I: Maternal weight gain in pregnancy is positively associated with fetal AC in second trimester. This study adds information that fetal growth regulation can be traced already in second trimester. Study II and III: The use of conditional centiles for EFW and CPR in combination with conventional centiles improved prediction of adverse perinatal outcomes. The results support an increased use of

conditional growth centiles in the monitoring of fetuses at risk, while the large individual physiological variation in CPR may limits the test performance. In general, our results indicate that there is merit in the further development of using serial observations to improve the prediction of adverse perinatal outcomes.

List of papers I-III

Paper I:

Hellebust H, Johnsen SL, Rasmussen S, Kiserud T. **Maternal weight gain: determinant for fetal abdominal circumference at second trimester.** *Acta Obstet Gynecol Scand* 2011 Jun: 90(6): 666-70.

Paper II:

Karlsen HO, Johnsen SL, Rasmussen S, Kiserud T. **Prediction of adverse neonatal outcomes using size centiles and conditional growth centiles.** *Ultrasound Obstet Gynecol* (2015 Dec: Epub ahead of print)

Paper III:

Karlsen HO, Ebbing C, Rasmussen S, Kiserud T, Johnsen SL. **Use of conditional centiles of middle cerebral artery pulsatility index and cerebroplacental ratio in the prediction of adverse perinatal outcomes.** (Revised version submitted in March 2016, *Acta Obstet Gynecol Scand*)

1 Introduction

1.1 Prenatal care in a historical perspective

Fetal size at birth and the enigma of fetal development has always been an interesting topic. An increased systematic measurement and registration of birthweight during the last century, has contributed to an increased understanding of fetal growth and fetal development. A photo of a 100 years old baby weigher is shown in Figure 1. Fetal size and growth has now become one of the main focuses in pregnancy care. During the first half of the 20th century knowledge of fetal development and factors influencing birthweight was based on examination after abortions and birth (1, 2). At this time the majority of births took place at home (3). From 1920 to 1960 there was an extensive increase in the number of birth institutions and women increasingly chose an institutionally delivery (3). Birthweight was recorded routinely for neonates born at St. Helens Hospital, New Zealand prior to 1922 (4) and institutionally births were recorded in similar ways in other countries. For those who had a delivery outside an institution there is no information on organized registration of medical information in birth records in the early 1900s.

Figure 1. *Hughes' baby weigher no 48B. Private photo.*

Organized antenatal care was virtually absent in the early 1900s. It was the fight against maternal and perinatal mortality that attracted the Norwegian doctors to the antenatal period, and led to a proposal of organized antenatal care in the 1930s (5). In

Norway a systematic registration of all birth data started when the Medical Birth Registry (MBR) was established in 1967. The first report revealed a higher perinatal mortality rate in Norway than in other Nordic countries (6), this prompted an increased focus on antenatal, perinatal and neonatal care. Specific recommendations concerning pregnancy care were set and perinatal audit committees were established (7).

Assessment of fetal size by abdominal palpation has largely been replaced by measurement of the fundal height, i.e. the symphysis-fundus (SF) measurement which was introduced in Stockholm, Sweden in 1972 by Westin (8). Both methods have low sensitivity in detection of small for gestational age (SGA) fetuses (9-12), which commonly is defined by an estimated fetal weight (EFW) $<10^{th}$ or $<5^{th}$ centile for gestational age. Abdominal palpation and SF height were the only antenatal methods available to identify impaired fetal growth until ultrasound imaging was introduced. In 1958 Ian Donald established the potential use of this technology (13), and in 1961 Donald and Brown introduced the measurement of biparietal diameter (BPD) (14), which later was shown to correlate with fetal weight (15). Based on a consensus conference routine ultrasound scan in the second trimester was introduced in Norway in 1986. The introduction of ultrasound in obstetric care has provided us with a unique opportunity to monitor fetal development and growth. Reference charts for fetal size and growth has regularly been updated in line with increasing knowledge and technical development. Identification of fetal growth restriction has become more detailed during the latest decades. Different methods are used in management of these pregnancies, except for repeated biometry measurements, surveillance are now supplemented by Doppler ultrasound and cardiotocography (CTG) registration. This rapid development in antenatal care has been reserved for industrialized countries; developing countries have partly still conditions comparable to that existed here in the early 1900s. In these countries there is also a big difference in availability of health care depending on whether you are rich or poor.

1.2 Gestational age

Information on gestational age is essential in pregnancy care. All reference charts e.g. for SF height, ultrasound estimates of fetal size and Doppler measurements are gestational age dependent. When it comes to ethical difficult decisions such as second trimester termination of pregnancy and delivery of extremely preterm fetuses, a correct gestational age is crucial. In post-term pregnancies a correct gestational age is also of great importance, since the risk of adverse perinatal outcomes, including intrauterine fetal death, increases if the pregnancy lasts beyond 294 days (16-18). Historically the expected date of delivery was based on Naegele's rule, where 280 days were added to the first day of the woman's last menstrual period (LMP). In a global perspective, this is the most common method. Prerequisites for using this method are correct recall of the first day of LMP, regular menstrual cycle, ovulation 14 days before next expected menstrual period and no use of hormonal contraceptives the past three months. However, these criteria are commonly not met and the gestational age assessment rendered correspondingly unreliable (19, 20). Ultrasound dating has proven to be more accurate compared to LMP dating, additionally it seems to reduce post term births (21-23). However, fetal growth is assumed to be under biological variation already at the earliest stages of pregnancy, and the variation increases with gestation. This implies that the accuracy of ultrasound dating, which relies on fetal biometry, reduces as the pregnancy progresses (24-26). At the second trimester scan at gestational week 17-20 the variability is as high as ± 7-10 days (2 standard deviations (SD)) (26, 27). The American College of Obstetricians and Gynaecologists suggest use of the earliest biometry measurement to determine gestational age (27), and this approach is supported by the majority of international societies of obstetrics (28), including the Norwegian Association of Obstetricians and Gynaecologist. A crown-rump length (CRL) measurement between gestational week 6 to 14 was introduced by Robinson in 1975 (29). The method is still considered to be the most accurate (20, 22, 25, 30) with a variability of ±3-5 days (2SD) (29, 31, 32). From 12 weeks onward fetal head circumference (HC) or BPD is preferred over CRL (32). For in vitro fertilized (IVF) pregnancies the day of conception is known, and there is therefore a broad consensus

that gestational age should not be adjusted with ultrasound in these pregnancies (28, 33)

Normal duration of a pregnancy is defined to be 280 days (range 259 – 294 days) by World Health Organization (WHO) (34). However, population based birth registries based on LMP suggest a longer pregnancy duration, the median day of delivery varied between 282 and 284 days (35-37). A median pregnancy duration of 282 days are now commonly used (35). The probability of having a spontaneous start of labor within 7 days of the estimated date of delivery (EDD) is about 60%, and the overwhelming majority of births are distributed during a five weeks period that lasts from gestational age 37 to 42 weeks (37). Fetal size in the second trimester is shown to be a determinant of pregnancy duration. Fetuses that had a smaller abdominal circumference (AC) than expected in the second trimester tend to have a longer pregnancy than fetuses with a larger AC (38). The effect being significant also after adjustment for the discrepancy between gestational ages based on LMP and that on ultrasound biometry of HC.

1.3 Normal fetal growth

Birthweight results from a complex interaction between genetic, environmental and maternoplacental factors (39-42). The normal biological variation is wide and increases throughout pregnancy, a fact, which is evident looking at the wide range of normal size of neonates at birth (3015-4140 gram referred to the 10^{th} and 90^{th} centile at 40 gestational weeks for female neonates) (43).

Fetal development in first trimester is dominated by organogenesis, and the nutritional supply of the embryo is provided by the yolk sac, until the placental circulation is established in the late first trimester (44). The CRL is commonly used to determine fetal size in the first trimester and first trimester fetal growth is associated with birthweight (45, 46). In the second and third trimester the fetus has increasing needs of nutrients and oxygen, which are provided by the placenta. The fetal genome, maternal health and nutritional supply by the placenta will be crucial for setting the fetal growth

trajectory. These factors will have increasing influence during pregnancy and contributes to the wide normal range in fetal size at birth. Reference charts for fetal size and growth demonstrate this wide variation well and is a helpful tool in identifying SGA and intrauterine growth restriction (IUGR) (47).

1.4 Methods

Repeated measurements of SF height are routinely used to assess fetal growth in antenatal care (8, 48-50). The finding of low or stationary SF heights, usually leads to referral for ultrasound estimation of fetal size. However, the sensitivity in detecting SGA fetuses by SF height is highly variable (14-76%) and in many studies reported to be low (49-51). Clinicians should be aware of a high false-negative rate for SGA identification. Calculation of EFW by ultrasound is based on a combination of different biometry measurements like femur length (FL), AC or mean abdominal diameter (MAD) and HC or BPD. A diversity of commonly used formulas is available for calculation of EFW such as Combs, Hadlock, Mielke and Dudley's formulas (52-55). The accuracy of EFW in prediction of birthweight has been reported to have a mean absolute prediction error between 7.5% and 18.8% (56). Formulas including head, abdomen and femur measurements showed lowest mean absolute error (56, 57). In large for gestational age (LGA) fetuses and in IUGR fetuses the accuracy is lower, with a tendency to overestimate fetal size in IUGR fetuses (58, 59) and underestimate size in large fetuses (57, 60). Maternal obesity and oligohydramnios influence sonographic insonation quality, but accuracy in fetal weight estimation has not shown to be significantly affected by maternal body mass index (BMI) (61, 62), while results are conflicting for oligohydramnios (63-65).

To assess fetal size and growth, appropriate reference ranges should be used (66). Cross sectional reference ranges are suitable for the assessment of fetal size at a given gestational age, but are poorly suited to assess growth. Growth is change in size over a period of time. To assess fetal growth longitudinal reference ranges based on serial observations are most appropriate (47, 67-70). One method of quantifying growth would be the calculation of conditional growth centiles, i.e. a previous measurement of

size in an individual fetus is utilized to establish the prediction range for the next measurement in that particular fetus (47, 67). These ranges are narrower compared with the reference ranges for the entire population and shifted toward the initial size centile. Examples are shown in Figure 2 and 3.

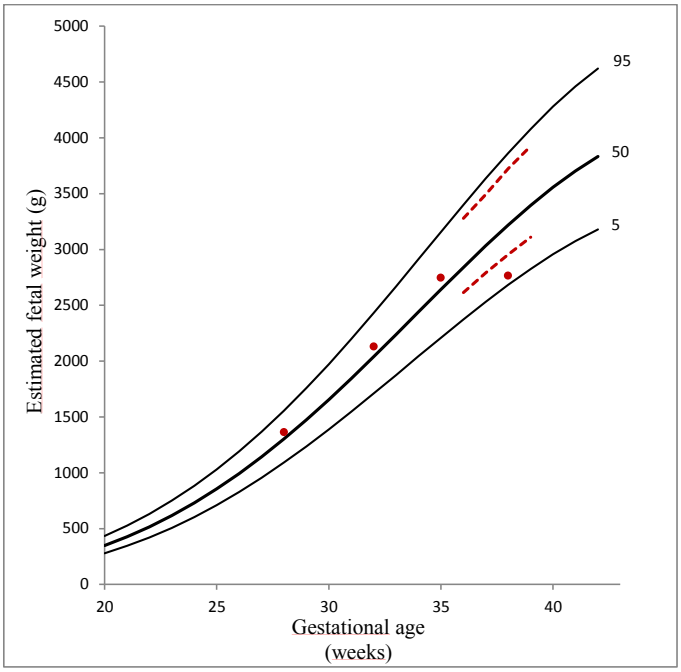


Figure 2. *Serial biometry measurements of estimated fetal weight (EFW) (red dots) plotted on reference ranges for male fetal sex with 5th, 50th and 95th centiles (black rules). The second last EFW assessment at 35 weeks (2746g, size centile 64) was used for calculating the individual conditional growth range for the last measurement at 38 weeks (red broken rules for 5th and 95th conditional growth centiles). The last EFW of 2760g corresponds to 11th centile for size and the conditional growth centile of 1.*

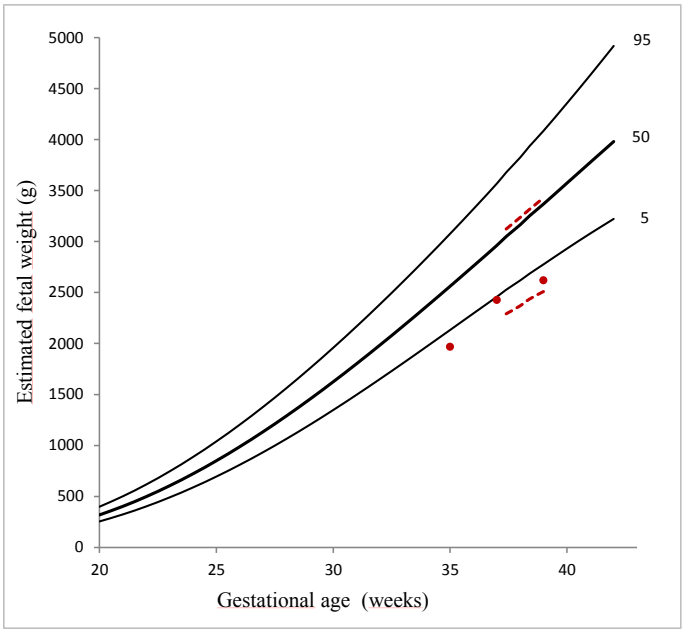


Figure 3. *Serial biometry measurements of estimated fetal weight (EFW) (red dots) plotted on reference ranges for male fetal sex with 5th, 50th and 95th centiles (black rules). The second last EFW assessment at 37 weeks (2427g, size centile 5) was used for calculating the individual conditional growth range for the last measurement at 39 weeks (red broken rules for 5th and 95th conditional growth centiles). The last EFW of 2618g corresponds to 3rd centile for size and the conditional growth centile of 11.*

1.5 Maternal influence on fetal size and growth

Fetal growth and fetal size at birth is mainly determined by the interaction of the fetal genome and maternal constraint (39, 71), both contributes to the wide range of normal biological variation in fetal size (43, 47).

1.5.1 Genetic influence on birthweight

Genetic influence on birthweight has mainly been described in epidemiological studies. In a study from the Swedish Twin and Birth Registers the heritability for birthweight in offspring of twins was estimated to 25-40% (72). Such registers provides a unique opportunity to perform intergenerational studies. Parent–offspring data from the MBR of Norway were used to analyze genetic influence of the normal variation of birthweight. Fetal genetic factors were estimated to explain 31% of the normal variation in birthweight, while maternal genetic factors explained 22% of the

variation (73). The variation in estimates of genetic contribution is large; it varies between 38-80% (39, 74). The mechanisms of interaction between genetic, maternal and environmental influences on fetal growth are not fully exploited.

1.5.2 Maternal constraint

Maternal constraint involves maternal factors affecting fetal growth through limited access to nutrients, metabolic and hormonal processes (71). Maternal anthropometric and nutritional status limits the nutritional capacity, but maternoplacental function is crucial for the final nutrient supply to the fetus. Several hormones are important for placental diffusion capacity, i.e. placental growth hormone and placental lactogen, both contributing to insulin resistance (75). Maternal constraint is regarded as a physiological process that is present in all pregnancies but to varying degree. In cases where fetal demand of nutrients exceeds the supply, maternal constraint can result in slow fetal growth with consequences for both neonatal and adult health. A recent study of 1 mill pregnancies showed that the 80-84th birthweight centiles had the lowest perinatal mortality (76). The authors interpreted their results in an evolutionary perspective, that maternal constrain taking care of maternal survival restricts fetal growth beyond optimal weight for offspring survival.

1.5.3 Maternal anthropometric measures

Maternal size and body composition influence fetal growth and proportions throughout the pregnancy. In first trimester low maternal stature is associated to lower CRL measurement (77), while several studies agree that pre-pregnant BMI has no influence on CRL (78, 79). During second half of pregnancy maternal height, pre pregnant weight and BMI influence fetal size with increasing effect (80-82). Most studies evaluate the effect on fetal size comparing pregnant women with low vs. high stature, weight and BMI, while cut offs varies between the studies. However, there is an agreement on a positive association between EFW and maternal stature and BMI (81, 82). Different maternal anthropometric measures influence biometric parameters at varying degree and at different stages in pregnancy. Goldenberg et al. found that low stature (<157 cm) vs. high stature (≥167 cm) affected fetal HC from week 31 and onwards, FL from week 25 and AC from week 36. While low BMI (<19.5) vs high

BMI (≥26) first influenced AC at week 25, followed by HC from week 31, and finally FL at week 36 (82). The study population in this study consisted of a high percentage of smokers (49%) and women with non-Caucasian ethnicity (69%). Therefore these results cannot be generalized to Norwegian pregnant women.

1.5.4 Gestational weight gain

In 2009 the Institute of Medicine in USA published new guidelines for maternal weight gain in pregnancy based on optimal maternal and fetal outcomes (83). Recommended weight gain differs according to pre-pregnant BMI categories: overweight or obese women should gain less weight (7.0-11.5 and 5.0-9.0 kg, respectively) than women with normal BMI (11.5-16.0 kg), while the opposite is recommended for underweight women (12.5-18.0 kg). Gestational weight gain reflects growth of maternal tissue and fetal growth, but also caloric intake during pregnancy.

A positive relation between gestational weight gain and birthweight are well documented (81, 84-86). Maternal weight gain below the recommended range is associated with low birthweight and excessive weight gain increases the risk of having a LGA neonate (81, 87-89). Several studies have shown that maternal weight gain during the 2nd trimester has the greatest impact on birthweight (85, 86, 88). There has been less focus on when in pregnancy fetal growth is influenced. Only one study has explored the relationship between gestational weight gain and biometric parameters at different gestational age (82). Goldenberg et al. found a significant lower fetal AC from gestational week 25 onwards in women with low total weight gain (<8 kg) compared to those with high total weight gain (≥16 kg). HC was first affected in gestational week 31 and FL from week 36 and no effect on biometric parameters was seen in week 18.

The area which the fetal AC measurement covers includes the stomach, subcutaneous and intra-abdominal fat accretion but is dominated by liver tissue. The mechanism in which gestational weight gain influence fetal AC is not known, but it has been shown that low maternal weight gain is associated with reduced umbilical venous perfusion to the right lobe of the fetal liver (90). This altered flow distribution may influence fetal

liver proliferation and production of insulin-like growth factor (IgF) 1 and 2 and as a consequence fetal growth will be affected (91, 92).

1.5.5 Other lifestyle related factors influencing fetal growth

Cigarette smoking during pregnancy is well known to reduce birthweight, and a dose response effect is evident (93-95). Alcohol consumption in pregnancy is also inversely related to birthweight but not to the extent of smoking (96-98). A meta-analysis has shown that moderate physical activity during pregnancy reduces the risk of having a LGA neonate without increasing the risk of having a SGA neonate (99). A positive association between maternal education and birthweight is seen (97).Education is thought to be an indirect parameter influencing fetal growth, higher education is related to other factors that have positive effect on fetal growth such as higher maternal age and less smoking (97).

1.6 Fetal growth restriction

Suspected IUGR is a common issue in antenatal care mounting to 3-7% of all pregnancies. IUGR is a major contributor to perinatal morbidity and mortality (100-102), in addition the birthweight gradient across the entire population is inversely linked to increased risk of adult diseases such as diabetes and cardiovascular diseases (103). For the immediate perinatal outcomes the identification, close monitoring and timely delivery are key aspects in optimizing management. This is still a huge challenge in developing countries where the pregnant population in general is less healthy and where the availability of qualified personnel and technology are limited.

1.6.1 Definition

Fetal growth restriction has many denominations and at least as many definitions. Numerous publications on this topic exist but the inconsistency in definitions contributes to some confusion and difficulties in comparing different studies. The definition of the WHO of low birthweight being <2500 g and for very low birthweight <1500 g is useful because it classifies a group neonates with high perinatal morbidity in societies where accurate gestational age is commonly unknown (34). However, the uncertainty of gestational age is also an important limitation when trying to

discriminate prematurity and growth restriction as the cause of morbidity in these societies.

Originally the term SGA was used to describe neonatal size at birth, but this term is also commonly used to describe intrauterine fetal size, with different cut offs 10^{th}, 5^{th} or 3^{rd} centile. A low cut off will more accurately identify the true growth restricted neonates but some normally sized neonates with impaired growth will be overlooked. Use of a high cut off will include numerous constitutionally small but well growing fetuses.

Many different reference curves for intrauterine fetal size and birthweight exist (43, 104). Whether to use a reference curve based on intrauterine observations or to use birthweight curves to identify SGA is debated. Population based reference curves of birthweight differ considerably from intrauterine reference curves especially at low gestations (43, 105, 106). Births at low gestation are often associated with pregnancy complications, which influence fetal growth; there is an increased incidence of IUGR in these pregnancies. The 10^{th} centile tends to be shifted lower in population based reference curves of birthweight than in intrauterine reference curves based on ultrasound EFW; as a consequence IUGR will be underdiagnosed.

The IUGR definition is intended to describe intrauterine growth conditions. Unfortunately, a lot of different definitions of IUGR exist (107). The most common definition is EFW $<10^{th}$ centile (27, 108), although, lower cut offs have shown to be better predictors of adverse perinatal outcomes (109). Other widely used descriptions are EFW <2SD (105) and fetal AC $<10^{th}$ centile or <2SD (108). A low fetal AC centile in combination with normal HC parameter is termed asymmetrical growth (110) while there is conflicting results regarding a HC/AC ratio above 95^{th} centile is associated to increased risk of adverse outcomes (111, 112). One single measurement of fetal size is not suitable to describe intrauterine growth (113). Serial ultrasound measurements and calculation of fetal growth expressed in gram/week or in conditional centiles (67), can

be used to discriminate between well growing small fetuses and those who have impaired intrauterine growth.

1.6.2 Etiology of fetal growth restriction

Although the mechanisms leading to fetal growth restriction are not completely understood, a number of important factors are known. Some prominent factors in the maternoplacental circulation and fetal and maternal conditions are described here. A large proportion of IUGR is due to placental dysfunction as seen in hypertensive pregnancy disorders (114, 115). Early onset IUGR (onset before 34 weeks gestation) is associated with pre-eclampsia (PE) in up to 50% of cases and account for 20-30% of all IUGR cases (116). Late onset IUGR is associated to PE in approximately 10% of the cases (116). Marginal or velamentous insertion of the umbilical cord on the placenta and a single umbilical artery are also associated with slow fetal growth (117). Uterine malformations, bicornuate uterus or uterus didelphys, predispose to slow fetal growth (118). Maternal diseases such as chronic hypertension, renal failure, rheumatic disease, pre gestational diabetes and eating disorders are closely associated with fetal growth restriction (119-127). Exposure to smoking or alcohol during pregnancy can affect fetal growth (128). Fetal causes of IUGR such as chromosomal aberrations, fetal anomalies (129-131) and intrauterine fetal infections (132) are associated with a less favorable prognosis compared to IUGR due to circumstances outside the fetus. If an obvious explanation of SGA cannot be found and the fetus grows within normal ranges, it is probably a constitutionally healthy small fetus.

1.6.3 Surveillance of IUGR

As IUGR fetuses are at risk of perinatal morbidity and mortality, increased surveillance during pregnancy and birth is required, compared to the surveillance applied to the general pregnant population. Management of a pregnancy with IUGR depends on gestational age. The key aspect in management is to find the optimal time and method for delivery based on a balance between the risk of fetal harm induced by leaving the fetus in utero and the risk of morbidity caused by iatrogenic prematurity. Surveillance includes repeated ultrasound examinations including fetal biometry and biophysical profile (amniotic fluid index, fetal movements and fetal heart rate

monitoring). A two-week interval between two ultrasound estimations of fetal weight is common in clinical practice, but an interval of three weeks is recommended to minimize the false positive rate of diagnosing fetal growth restriction (133). Early onset IURG (<34 weeks gestation) is considered to be more severe than late onset IUGR and fetal condition may be followed more closely. Decelerated fetal growth is usually accompanied by circulatory redistribution to protect the fetus against hypoxic damage (134, 135). Prioritized organs include the fetal brain, heart and adrenal glands (136). The introduction of fetal Doppler ultrasound has given us a unique opportunity to study the human circulation in utero and monitor fetuses at risk.

1.7 Doppler ultrasound assessment of placental and fetal circulation

Doppler ultrasound was introduced in obstetrics before 1980 and has since been developed to a range of techniques now widely in use to study fetal circulation (Figure 4). The fetus receives well-oxygenated blood from the placenta through the umbilical vein and deoxygenated blood is directed from the fetus to the placenta through the umbilical arteries (UA). At mid-gestation approximately one half of the fetal total blood volume is located in the placenta; it gradually decreases to 25-30% at term (137). The fetus is capable of fast redistribution of blood if needed, three fetal shunts (ductus venosus (DV), foramen ovale and ductus arteriosus) contribute to distribution of oxygenated blood to prioritized organs when needed (135). This flexible circulatory system is extensively studied by Doppler ultrasound and reference ranges are established under physiological conditions in human fetuses (138-142). Correspondingly, the pattern of circulatory changes in growth restricted fetuses due to placental dysfunction are reasonably well described (143-146) and used successfully to identify those who are at the highest risk of adverse outcomes (147-149).

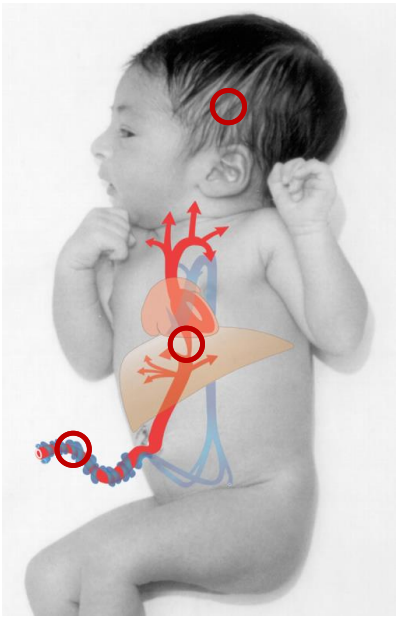


Figure 4. *Demonstration of central fetal circulation (reproduced by permission (150). The most common vessels being examined by Doppler are the umbilical artery, Ductus venosus and middle cerebral artery; sites are marked by red circles.*

1.7.1 Maternoplacental circulation

The uterine arteries (UtA) are the main contributors to uterine blood flow which is increasing tenfold during pregnancy (151, 152). This increase in blood flow is mediated by placental trophoblastic invasion of the spiral arteries (153, 154). The process starts around gestational week 8 and the transformation of the uterine spiral arteries from a high resistant vascular system to a low resistant vascular system is completed around week 24 (153, 154). Campbell et al. introduced use of Doppler sonography of the UtA in 1983 after their discovery of an association between increased vascular resistance and pregnancy complications such as PE and poor fetal growth (155). Impaired trophoblastic invasion may cause increase UtA resistance, which can be measured by Doppler ultrasound and be traced as high pulsatility index (PI). Such a finding is associated to IUGR, PE, placental abruption and stillbirth (156-159). Reference ranges for UtA PI now exists from gestational age 11 to week 41 (138). In pregnancies at risk of PE and IUGR an evaluation of UtA PI has traditionally been performed in gestational week 23-24 (160). Recent studies have also

demonstrated that an examination of UtA in late first trimester also can be used to identify pregnant women at risk (161). In a low risk population such screening has limited benefit (161), while in a high risk population an evaluation of the maternoplacental circulation can be helpful in further management and a high resistance can be the first sign of IUGR due to placental dysfunction.

1.7.2 The umbilical artery

Between gestational weeks 20 to 32 approximately one third of the fetal cardiac output is directed to the placenta via the UA (162, 163). After gestational week 32 this volume is gradually reduced to one fifth at term (162, 163). The vascular resistance in the placenta is mainly determined by the vascular bed, of which the area is increasing during pregnancy, resulting in reduced impedance. Endothelial cells in the placenta regulate angiogenesis and vasomotor tone. Normally the UA Doppler waveform is characterized by high diastolic blood flow velocity, while in cases of placental dysfunction abnormal angiogenesis and increased vasoconstriction results in increased vascular resistance in the placenta and the diastolic blood flow velocity is reduced (164). An increased flow resistant in the UA is associated with SGA due to placental dysfunction (165), while a PI >95th centile will not be evident before at least 30% of the placenta is affected (166, 167). A normal UA Doppler measurement is therefore not sufficient to assess fetal wellbeing (168, 169). In severe cases absent end diastolic (AED) blood flow can be seen and by further deterioration absent and reversed end diastolic (ARED) blood flow can occur (166, 167, 170). When 60-70% of the placental villi are destructed AED or ARED typically occur and the risk of perinatal morbidity and mortality is high (166, 170, 171). The mortality rate varies between 5 to 36% and is dependent on gestational age and the degree of affection of the venous fetal circulation (170, 171). AED and ARED blood flow in the UA are mainly seen in early onset IUGR, it is an uncommon finding late in pregnancy. UA Doppler waveform analysis is now an integrated part of surveillance in high risk pregnancies and are widely used in fetuses <10th centile to identify those at increased risk of adverse outcomes (171-173).

1.7.3 The blood flow of the middle cerebral artery

Normally 15% of the cardiac output is directed to the fetal brain. In cases of reduced placental perfusion, hypoxia, increase in pCO_2 or low arterial pH, the fetus can increase its blood supply to the brain (174, 175). This redistribution is called "brain sparing" and is characterized by vasodilatation of cerebral arteries resulting in an increased diastolic blood flow velocity and reduced resistance. Doppler recordings in the middle cerebral artery (MCA) are now an integrated part in the surveillance of fetal growth restriction. A low PI reflects redistribution of fetal cardiac output to the brain, and this brain sparing effect is associated to adverse perinatal outcomes (134, 176, 177).

Both the UA PI and MCA PI have an independent predictive effect on neonatal outcomes but recent studies have shown a better prediction of adverse outcomes combining these two in the cerebroplacental ratio (CPR) (178-180). The CPR reflects both the placental function and the fetal response to the placental return. However, the accuracy in identification of the fetuses at risk using CPR $<5^{th}$ centile varies between different studies, sensitivity ranged from 42% to 85% (149, 181, 182). The different study designs partly explain this wide range in sensitivity but may also be due to the wide physiological variation in MCA PI (140, 183), which also applies to CPR (140, 183). As for serial growth measurements, longitudinal reference chart for MCA PI and CPR are published and these allow calculation of conditional centiles for cerebral blood flow measurements (140). The use of conditional centiles in prediction of adverse perinatal outcomes is tested for fetal growth (184), while there are no publications regarding prediction of adverse perinatal outcomes by conditional centiles for fetal Doppler measurements. This way to exploit the value of serial measurements may be useful in prediction of adverse perinatal outcomes.

1.7.4 The Ductus Venosus

The fetus is supplied with oxygenated blood from the placenta via the umbilical vein. Blood from the umbilical vein is distributed to the liver and a fraction of 20-30% bypasses the liver through the DV into the heart (185). In placental insufficiency an increased fraction of well oxygenated blood from the umbilical vein is directed

through the DV to the fetal heart to ensure oxygenation of prioritized organs such as the brain, adrenal glands and heart (186, 187). Doppler measurement in the DV was introduced as a diagnostic tool in 1991(141). A relationship between the pulsatility index for veins (PIV) in the DV and the degree of acidemia has been demonstrated (188). Chronic hypoxia in the fetus causes myocardial dysfunction resulting in an increased afterload which can be seen as increased DV PIV. In early onset IUGR abnormal DV blood flow is considered a late and severe sign of fetal deterioration (143, 147, 189). In surveillance of these compromised fetuses DV blood flow is an important parameter in terms of delivery decisions (190, 191).

1.7.5 Circulatory redistribution in compromised fetuses

In an experimental animal study of fetal sheep's DV was blocked resulting in increased liver blood flow. Compared to the controls, cell proliferation was significantly increased in the liver and organs such as the heart, kidneys and skeletal muscle (192). The fetal liver is a key organ in regulation of fetal growth (91). Umbilical venous (UV) perfusion of the liver is essential for fetal growth due to the high content of nutrients. Of the total UV blood flow 70-80% is directed to the fetal liver (185). UV blood flow accounts for approximately 85% of the total venous liver perfusion, while the portal vein contributes with about 15%.

Liver perfusion is influenced by maternal body composition, weight gain in pregnancy and diet (90, 150). Slim mothers and those having an unbalanced diet had a compensatory increased liver blood flow (150), while low gestational weight gain was associated with reduced blood flow from the UV to the fetal liver (90). A similar pattern is seen in fetal growth restriction due to placental insufficiency (92, 186). In IUGR fetuses blood from the UV is redistributed away from the fetal liver to increase the amount of oxygenated blood to the fetal heart (143). In compromised fetuses a compensatory increase in portal blood flow to the right liver lobe will result in right liver lobe hypoxemia. These adaptive responses will affect liver growth and production of growth factors like IgF1 and 2 (91, 193). As a consequence liver size decreases, and since fetal AC is dominated by the fetal liver, a low AC measurement can be one of the first biometric signs of fetal growth restriction. Low IgF production

will further affect growth of other organs and this adaption to a suboptimal intrauterine environment may have longer term consequences for adult health (194).

Fetal response to hypoxia is redistribution of cardiac output and fetal compromise is associated with early arterial circulatory changes followed by alterations in the venous circulation. The sequence of the fetal deterioration is not uniform (147, 148). The fetal physiology changes during pregnancy and is evident looking at the different patterns of sequential changes in early onset IUGR. The first circulatory marker of IUGR due to placental disease is increased impedance in the UtA, which can be seen weeks before fetal circulation is affected (148). In early onset IUGR the most common sequence is abnormal UA PI, followed by low CPR and low PI in the MCA. Abnormal DV flow usually debuts after signs of brain sparing, but can also appear before brain sparing (147). Progressive changes in the UA such as AED and ARED or a PIV above the 95th centile in the DV are considered acute prognostic markers and the risk of acidosis is increased (148). Daily monitoring may be required and delivery must be considered with progressive fetal compromise, but this should be balanced against the risk of neonatal morbidity due to prematurity (195, 196). Other late signs in fetal deterioration are abnormal biophysical profile including reduced fetal movements, reduced breathing movements, oligohydramnios and abnormal CTG (low short term variation) (148). In a study of Baschat et al. such changes appeared one day prior to delivery (147). In late onset IUGR less severe circulatory changes are seen (190) and blood flow in the UA is not necessarily abnormal (177). Low CPR centile is often the first marker to be affected followed by a MCA PI <5 (146, 148), and monitoring intervals are to a large extent based on these parameters (195). Gestational age is of less importance in delivery decisions in late onset IUGR (195). In a randomized controlled trial of IUGR fetuses at term no difference in adverse outcomes was found between the induction group and those who were randomized to expectant management (197). However, any abnormal finding in the CPR, amniotic fluid index or CTG will generally lead to intervention.

1.7.6 Intrapartum care

Growth restricted fetuses may suffer from chronic hypoxia and they have limited capacity of further redistribution of the blood flow if they were exposed to acute hypoxia during labor (198). In early onset IUGR with affected venous circulation like abnormal DV blood flow, pulsations in the umbilical vein or in cases with AED or ARED flow in the UA, a caesarean delivery is the most common preferred route for delivery (191, 196). Fetal heart rate abnormalities appear more frequently in fetuses exposed to chronic hypoxia (199), and continuous intrapartum fetal heart rate monitoring is required (200, 201) to reveal any deterioration of hypoxia and to prevent neonatal asphyxia. A low centile of CPR increases the risk of intrapartum fetal distress and need for an emergency cesarean section, however the likelihood for vaginal birth exceeds 50% (168, 202, 203).

1.8 Consequences of IUGR

The increased risk of perinatal morbidity and mortality in IUGR fetuses is closely linked to prematurity; some studies have shown that fetal growth restriction in itself is an individual risk factor (204, 205). Several studies have evaluated the risk of neonatal morbidity and mortality. However, the impact of IUGR on neurodevelopment in childhood and long term consequences for adult health has gained increased interest in the latest decades (206).

1.8.1 Short term consequences

IUGR fetuses are highly susceptible to iatrogenic delivery and in cases of early onset IUGR the risk of prematurity is high. Early preterm birth is associated with mortality and severe morbidity (207). Late preterm birth is also associated with significant neonatal morbidity (208). The consequences of prematurity are further increased in IUGR fetuses compared to normally size preterm fetuses (101). In the TRUFFLE study a death rate of 5.5% and severe morbidity rate of 24% in early onset IUGR fetuses was found (191). IUGR fetuses are exposed to an increased risk of intrauterine fetal death, and it has been shown that the risk increases with gestational age and is inversely related to birthweight centiles (209). Late onset IUGR is more often undetected and accounts for over 50% of unanticipated intrauterine fetal deaths (210).

In a recent study the combination of EFW <10th centile and slow growth of the fetal AC showed the highest risk of adverse outcomes like metabolic acidosis, Apgar score <7 after 5 minutes and admission to NICU in term neonates (211).

Placental compromise is a common cause of IUGR and results in reduction of nutrients and oxygen delivery, and circulatory redistribution will eventually appear. As a result the changes make these fetuses more susceptible to hypoxia during uterine contractions in labor; the contractions will lead to additional need for redistribution (212). Signs of hypoxia during labor like fetal heart rate decelerations entail an increased need for emergency caesarean delivery in IUGR fetuses (148, 203). The second stage of labor is usually the most stressful part of the delivery for the fetus (213, 214), but surprisingly few studies have examined if an increased risk of operative vaginal delivery due to fetal distress is present in IUGR fetuses, compared to AGA fetuses.

Markers of perinatal asphyxia such as low 5 minutes Apgar score (<7) and metabolic acidosis have different prognostic value. IUGR fetuses have significantly increased risk of low Apgar score compared to AGA neonates (215). However, the association with impaired neurologic development is modest for a 5 minute Apgar score <7. While a 5 minutes Apgar score <4 is a stronger predictor of neurological sequelae (216). Metabolic acidosis (umbilical artery pH <7.05 and base deficit >12.0 mmol/L) is a more objective parameter in the evaluation of fetal well-being immediately after birth. This parameter is one of the criteria for intrapartum asphyxia, which is associated with neonatal morbidity and mortality (217-219). An increased risk of neurologic impairment at 6.5 years of age is demonstrated, but the risk is mainly reserved for those who had other clinical signs of asphyxia or encephalopathy in the neonatal period (220). Growth restricted neonates are also at risk of having hypoglycemia shortly after birth (221), undiagnosed and untreated this can potentially lead to neurologic damage with long term consequences (222).

1.8.2 Long term consequences

One of the most feared complications following delivery is impaired neurodevelopment. The prognosis can be difficult to predict in the neonatal period as the problems may occur several years later and range from mild learning problems and mild hyperactivity disorder to severe cerebral palsy. The severity of prematurity is the most important factor for neurologic outcome (223, 224), and the addition of IUGR in these fetuses has not shown to have an independent predictive effect on neurological outcomes (224, 225). While in cases of late preterm and term deliveries SGA neonates have shown a significantly lower intelligence score, neurodevelopment score and school achievements compared to age-matched controls (226-228). Perinatal asphyxia is a well-known risk factor for impaired neurodevelopment, while the presence of metabolic acidosis at birth is associated to adverse neurologic outcome only in the presence of symptoms of encephalopathy (220). Two randomized trials have evaluated neurodevelopment in infants at 2 years of age in preterm growth restricted neonates (229, 230). None of the studies shown reduced neurodevelopment outcomes in the groups who were randomized to delayed delivery, which supports the importance of achieving as high gestational age as possible to reduce the risk of other severe outcomes associated to prematurity.

The awareness of increased rates of cardiovascular disease and type II diabetes in adults born with low birthweight led to the Barker hypothesis (194, 231). This theory of developmental origins of adult disease has gradually gained acceptance, and the knowledge of how the fetus adapt to a suboptimal intrauterine environment and undernutrition is increasing. Intrauterine programming is associated to changes in both organ structure and function with potentially consequences for extra-uterine life (91, 232, 233). In a Swedish cohort slow fetal growth was associated with increased risk of death from ischemic heart disease (234). An inverse relationship between birthweight and systolic blood pressure from adolescence and onwards has been demonstrated (235). Hypertension is one of the medical conditions included in the metabolic syndrome in addition to reduced glucose tolerance, abdominal obesity and hyperlipidemia. Low birthweight has also been associated with the metabolic

syndrome in young adulthood and onwards (236). Additionally, osteoporosis has been linked to impaired intrauterine nutrition (237, 238).

1.9 Ultrasound physics

Ultrasound waves have frequencies above the audible range (>20 kHz). Diagnostic ultrasound in obstetrics and gynecology normally operates in the range of 2-10 MHz. Low frequencies probes provide better penetration of the tissues but lower resolution, while high frequencies probes provide better resolution and are therefore most suitable for imaging of superficial structures. Resolution refers to the smallest distance between two spots that can be visually discriminated.

Brightness mode (B-mode) scanning is used for making two dimensional grey scale images. Sound waves produced by an ultrasound transducer are passed through different tissues, being absorbed, scattered and reflected in different degree depending on the density of the tissue. Reflected waves are called an echo, and the echoes captured by the transducer are used to generate an ultrasound image. Two-dimensional grey scale ultrasound is widely used in evaluation and measurements of fetal structures.

Doppler ultrasound is increasingly used in obstetric settings to monitor risk pregnancies by evaluation of blood flow velocities. The Doppler Effect is change in frequency of an ultrasound wave that is detected by an observer which moves relative to the wave source. This frequency change is called the Doppler shift. Different types of Doppler ultrasound are used in clinical practice, continuous-wave Doppler, color Doppler, power Doppler and pulsed Doppler.

- Continuous wave Doppler ultrasound is a technique in which the transducer emits and receives ultrasound waves continuously. It is widely used for external monitoring of the fetal heart.
- Color Doppler ultrasound is a form of pulse wave Doppler where the measured Doppler shift is transformed into an assigned colour depending on the flow direction. The color display is superimposed on the grey scale image.

- Power Doppler ultrasound is based on the amplitude, or power, of Doppler signals, reflecting the number of red blood cells scattering the ultrasonic beam. This allows detection of low velocity flow and a better visualization of small vessels, but at the expense of information on velocity and direction.
- In the pulse wave Doppler ultrasound the transducer emits ultrasound in repeated pulses at a given pulse repetition frequency. This technique contributes with information about the blood velocity profile. In velocity measurements an optimal insonation angle (<30°) is important to avoid a false low velocity.

1.10 Ultrasound safety

Safety in obstetric ultrasound has been subject for increased focus in the last decade. Ultrasound scanners produced today have a maximal permitted intensity of 720 mW/cm^2 spatial peak temporal average (SPTA), while prior to 1991 the maximum limit was 94 mW/cm^2 SPTA. This increase in intensity gave improved image quality but also potentially increased risk of harmful effects. So far there is no evidence that diagnostic ultrasound has harmful effects on the developing human fetus. An effect on birthweight (239, 240), neurological development (241-245) and malignancy (246, 247) has been in focus, without demonstrated consequence. The only documented bio effect of prenatal grey scale ultrasound is non-right handedness in male neonates (244, 248). Doppler ultrasound represents increased output energy compared to B-mode scanning. The growing use of Doppler ultrasound has caused increased concern and research regarding potentially harmful effects (249).

Ultrasound imaging deposits energy in the body in terms of increased temperature and mechanical cavitation. From 1993 information about these two indices, thermal index (TI) and mechanical index (MI) have been provided with all imaging machines and this information are visible on the display of the machines. In this way the operator can prevent unnecessary use of potentially harmful high output energy. The British Medical Ultrasound Society (BMUS) supports the ALARA (i.e. as low as reasonable achievable) principle for safe use of ultrasound energy output. Sande et al. showed that reducing the energy output from thermal index for bone (TIB) from 1.0 to 0.1 does

neither compromise the ultrasound biometry measurements (250) nor the Doppler measurements (251).

2 Aims of the studies

Study I

To assess whether maternal weight gain during pregnancy might influence fetal AC as early as second trimester in low risk pregnancies.

Study II

To test whether adding conditional growth centiles to centiles of estimated fetal weight improves the prediction of adverse perinatal outcomes compared with the SGA classification alone.

Study III

To test whether adding conditional centile for middle cerebral artery pulsatility index and cerebroplacental ratio to conventional centiles, improves the prediction of adverse perinatal outcomes compared to the use of conventional centiles alone.

3 Materials and methods

3.1 Study I

3.1.1 Study population

This study was a part of the "Fetal Age and Growth" project that included 650 women with a low risk pregnancy for the establishment of fetal size and growth charts (47, 80). Recruitment took place between August 2001 to September 2003 and they were included when they attended the routine ultrasound scan. All women participated voluntarily and gave their written informed consent according to The Regional Committee of Medical Research Ethics approval (REK-III no. 025.01). Inclusion criteria were healthy women with regular menstrual periods (28±4 days) in at least three months prior to this pregnancy and no use hormone therapy during these three months, a certain LMP date, singleton pregnancy, no history of complications in a previous pregnancy and no regular use of medication. Women with a discrepancy of ≥14 days between ultrasound and menstrual age were excluded.

3.1.2 Examinations

We collected information on maternal weight measurements from the antenatal forms. Weight gain during pregnancy was calculated as the difference between weight at the last antenatal visit and pre-pregnant weight. For the analysis we calculated weight gain per week, and created subgroups according to low weight gain (<0.28 kg per week), normal weight gain (0.28-0.40 kg per week) and high weight gain (>0.40 kg per week). Gestational age was based on LMP. The ultrasound examinations were performed by two experienced ultrasound operators, using a Philips HDI 5000 machine (Phillips Seattle, WA, USA), with a 2-5 MHz abdominal scanning probe, or Aloka Prosound-5000 machine (Aloka, Tokyo, Japan), with a 2-5 MHz abdominal scanning probe. Fetal AC ultrasound measurement was obtained using an ellipse in the transverse section of the fetal abdomen at the level where the umbilical vein enters the liver. The mean of three measurements at one of the visits between 15-25 weeks gestation was used.

3.1.3 Statistical method

Descriptive statistics were used to characterize the study population. Z-score for AC and weight gain per week were used to adjust for variation in gestational age at measurement. Linear regression analysis was used to assess the effect of maternal weight gain in pregnancy on AC and birth weight. Variables with a *p*-value <0.05 were considered statistically significant. We used SPSS (Statistical Package for the Social Sciences; Inc, Chicago, IL, USA) for the analysis.

3.2 Study II and III

3.2.1 Study population

During a four years period from May 2010 to June 2014, pregnant women were recruited to this prospective longitudinal study of fetal growth at Fetal Medicine Unit, Department of Obstetrics and Gynecology, Haukeland University Hospital, Bergen, Norway. The study was approved by the Regional Committees for Medical and Health Research Ethics (REC West no. 2010/686). Pregnant women referred for the 24-weeks ultrasound evaluation due to high risk of IUGR and pregnant women having had an ultrasound examination for any clinical indication and diagnosed with a SGA fetus ($\leq 5^{th}$ centile of EFW) were invited to participate in the study. Merely women with singleton pregnancies were invited to the study and all participants gave their written informed consent. Women were invited to participate due to high risk if they had previous PE and/or given birth to a SGA ($\leq 5^{th}$ centile) neonate, chronic maternal diseases as hypertension, renal failure, systemic rheumatic disease but women with pre-gestational diabetes were not included. Discrepancy of ≥14 days between the due dates set by LMP and ultrasound dating was also one of the inclusion criteria. Chromosomal aberrations or congenital malformations in the neonate were excluded.

3.2.2 Examinations

Gestational age was assessed by ultrasound of HC in second trimester (252) unless a 1^{st} trimester scan of CRL (29) had been carried out or if the day of conception was known due to IVF. Voluson 730 Expert, E6 or E8, GE Medical systems, Kretz

Ultrasound, Zipf Austria, were used for measurements of fetal biometry and Doppler ultrasound.

The ultrasound examinations included biometry measurements of the fetal HC, AC and FL (80). EFW was calculated according to Combs formula (52). The PI of the MCA and UA were measured at each visit and CPR was calculated by dividing the MCA PI with the UA PI. Examination was carried out with 2-6 weeks intervals and modified according to clinical needs. Participants where only one measurement was available were not included in the analysis. Size centile of EFW was calculated at each visit (47), and conditional growth centile was calculated between the last and the previous biometry with at least 14 days interval (47, 67, 69).

Decisions concerning timing of delivery and delivery mode were made by the clinicians in line with local and national guidelines. According to these guidelines SGA as an isolated finding was not indication for preterm delivery; these pregnancies were followed up until 39-40 weeks gestation as long as no additional factors appeared. Birth outcomes (gestational age, birthweight, information about labor and delivery, Apgar score and admission to neonatal intensive care unit (NICU)) were collected from medical records after birth. A glucose test of the neonate was taken within 2 hours after delivery. Preterm birth (<37 weeks gestation), operative delivery (including cesarean delivery and vaginal instrumental delivery) due to fetal distress, admission to the NICU, 5 minutes Apgar score <7, hypoglycemia (glucose <2.0 mmol/L), and perinatal mortality were considered adverse outcomes. A combined outcome variable “any adverse outcome” was established if one or more components were abnormal. Delivery due to fetal distress was indicated by pathological fetal Doppler findings, CTG abnormalities or due to fetal echocardiographic events (ST analysis, STAN) (201). Pregnancies at risk were monitored during labor by fetal echocardiography from gestational age 36 onwards. During the study period the overall cesarean rate in the department was 12.5%.

3.2.3 Statistical methods

In study II and III a power calculation was performed before the initiation of the study. This was based on Apgar score in a population of SGA neonates at or below the 5th centile born at Haukeland University Hospital in the period from January to August 2009. We aimed to show a difference in Apgar score of at least 2, with a significance level of 5% and a power of 90%. Using Altman's nomogram (253) we calculated a need of 44 participants in the SGA group. For the high risk group we had no direct power calculation, but recurrence rate of having an SGA (<5th centile) neonate is about 23% (254). Due to uncertainty about the power calculation we decided to perform an interim analysis after delivery of the first 80 participants using the 'any adverse outcome' variable as an outcome measure. We used the log-likelihood test to assess whether adding conditional growth centile ≤5 to a model with size ≤5th centile significantly improved the model (p <0.05) and estimated that a sample of 160 women was needed. To allow for potential withdrawals, exclusions, and incomplete data for some participants, the sample was expanded to 220 women.

We used log binomial regression analysis to assess whether size centiles and conditional growth centiles was associated with the outcomes, shown as Relative Risk (RR) with 95% confidence interval (CI). To test whether size and conditional growth centiles had independent association with the outcomes, when adjusted for each other, both parameters were included in the model and results were shown as adjusted Relative Risk (aRR) with 95% CI. Log-likelihood testing was used to test the hypothesis that conditional growth centile between the last and previous biometry, in combination with size centile at the last examination, improved the prediction of adverse outcomes compared with the use of fetal size centile alone.

Conditional centiles for MCA PI and CPR were calculated between the last measurement based on the penultimate measurement (67). The formula for conditional centiles includes gestational age, measurement and variance at the previous and current sessions in addition the covariance of both measurements. Log-binomial regression analysis was used to test the association between conditional centiles ≤5 or

≤10 for MCA PI and CPR at the final visit and adverse outcomes; the results are presented as RR with 95% CI. To test whether the centiles at the final visit and the corresponding conditional centiles ≤5 and ≤10 had independent association with the adverse outcomes, both the conventional and the conditional centiles were included in the model, and thereby adjusted for each other; the results are presented as aRR. In cases where both centiles had independent association with the outcomes, we were able to test the hypothesis that adding conditional centile to conventional centile for MCA PI and CPR improved the prediction of adverse outcomes compared with the use of conventional centile alone. Goodness of fit of the two models was compared using log-likelihood testing. To increase the sample size when log-binomial regression failed to show independent associations, all observations were used in multilevel log-binomial regression. Pairs of first to second, second to third, third to fourth, fourth to fifth, and fifth to sixth measurements were identified. We calculated conventional centiles and conditional centiles (5^{th} and 10^{th}) for the last measurement of CPR and MCA PI in each pair. Log-likelihood testing is not optimal in multilevel models. The possible improvement of the model adding conditional centiles to conventional centiles was instead assessed by change in Wald chi square between the models. To optimize these calculations we used Markov Chain Monte Carlo regression in the MLwiN program.

Diagnostic tests like positive predictive value (PPV), negative predictive value (NPV), sensitivity and specificity, were used to demonstrate the effect for the outcome variable 'any adverse outcome' with the $\leq 5^{th}$ centile as cut off for size and conditional growth centiles and the $\leq 10^{th}$ centile for CPR.

Possible collinearity between conventional and conditional centiles for both growth and Doppler measurements were assessed using variance inflation factor (VIF) (255). Statistical analyses were carried out using SPSS 22 (Statistical Package for the social Sciences, SPSS inc, Chicago, IL, USA) and a *p*-value <0.05 was considered significant.

4 Results

4.1 Study I

Complete data were obtained in 515 of the 650 women who were included. Information on the maternal characteristics of the study population is presented in table 1. Gestational age at the last antenatal visit ranged from 32^{+0} to 41^{+6}. Mean maternal weight gain per week was 0.39 kg. Two hundred and thirty women (45%) gained above 0.40 kg per week and 96 women (19%) gained less than 0.28 kg per week.

Table 1. Maternal characteristics of the study population (n=515)

Maternal characteristics	Median (range or %)
Pre-pregnancy weight (kg)	65 (43–132)
Weight gain (kg)	14 (5–34)
Height (cm)	168 (152–183)
BMI (kg/m^2)	23 (16–48)
Age (years)	30 (19–43)
Smokers (n)	41 (8%)

The 101 women with missing information of maternal weight at the last antenatal visit was comparable to the study population when it comes to median pre-pregnant weight (64 kg), height (169 cm) and BMI (23 kg/cm^2).

Using linear regression analysis we found a positive association between z-score for weekly maternal weight gain and z-score for second trimester fetal AC ($p = 0.001$). Subgroup analysis in the different weight gain categories showed a significant lower fetal AC measurement in those with low maternal weight gain ($p < 0.001$). Excess maternal weight gain (>0.40 kg per week), pre-pregnant weight and BMI were not associated to fetal AC in second trimester; results are presented in table 2.

Table 2. Effect of maternal pregnancy weight gain, pre-pregnant weight and BMI on fetal abdominal circumference at 15-25 weeks` gestation. R, 95% Cl and standard error (SE) expressed as z-score.

	n	r	SE	95% Cl	p-value
Weight gain	515	0.122	0.036	(0.051–0.194)	0.001
Weight gain>0.40 kg/week	230	0.020	0.091	(–0.160 to 0.199)	0.829
Weight gain<0.28 kg/week	96	0.554	0.147	(0.261–0.846)	<0.001
Pre-pregnancy weight	515	÷0.002	0.003	(–0.008 to 0.004)	0.483
Pre-pregnancy BMI	515	÷0.012	0.009	(–0.029 to 0.005)	0.171

4.2 Study II

Complete biometric data was available in 211 participants. Maternal characteristics and birth outcomes of the study population are shown in Table 3. Maternal characteristics and birth outcomes are also shown for those without any adverse outcomes (n = 122) and those with one or more adverse outcomes (n = 89) in table 3.

Table 3. Maternal characteristics and birth outcomes of the total study population, and in those without and those with adverse outcomes (*n*=211).

Characteristic	Median (range) or n (%)		
	Total study population (n = 211)	Non adverse outcomes (n = 122)	Adverse outcomes (n =89)
Maternal			
Age (years)	30 (17–43)	30 (21–42)	30 (17–43)
Height (cm)	165 (148–179)	165 (148–179)	164 (148–176)
Pre-pregnancy weight (kg)	63 (44–120)	62 (45–114)	65 (44–120)
BMI (kg/m^2)	22.9 (17.2–41.5)	22.6 (17.2–41.4)	24.1 (17.5–41.5)
Parity ≥1	174 (82.5%)	114 (93.4%)	60 (67.4%)
Smoking	17 (8.1%)	5 (4.1%)	12 (13.5%)
Chronic maternal disease	12 (5.7%)	4 (3.3%)	8 (9.0%)
Birth outcomes			
Gestational age at delivery ($weeks^{+days}$)	39^{+2} (25^{+3}–42^{+3})	40^{+0} (37^{+4}-$42^{3)}$	36^{+5} (25^{+3}–40^{+6})
Newborn birth weight (g)	2890 (440–4340)	3200 (2320–4340)	2270 (440–4135)
Birth weight ≤ 5^{th} centile	83 (39%)	27 (22.1%)	56 (62.9%)
Birth weight ≤ 10^{th} centile	108 (51%)	41 (33.6%)	67 (75.3%)
Newborn length (cm)	48 (28–54)	49 (43–54)	45 (28–53)
Ponderal index (kg/m^3)	26.1 (17.6–32.4)	27.0 (21.5–32.4)	24.5 (17.6–29.7)
Placenta weight (g)	500 (120–1100)	550 (300–1100)	425 (120–900)
Male infants	103 (49%)	60 (49.2%)	43 (48.3%)

Complete outcome data were available for all, except for 40 (19%) missing neonatal glucose levels. The frequency of the different perinatal outcomes is listed in Table 4. Both the perinatal deaths occurred in the neonatal period and severe growth restriction and extreme prematurity (<28 gestational weeks) were present in both cases.

Table 4. Perinatal outcomes in the total study population (n=211).

Outcomes	n (%)
Preterm birth (< 37 weeks)	50 (23.7%)
Operative delivery due to fetal distress	50 (23.7%)
Admission to NICU	48 (22.7%)
5 min Apgar <7	10 (4.7%)
Hypoglycemia (<2.0)	23 (10.9%)
Perinatal mortality	2 (0.9%)
'Any adverse outcome'	89 (42.2%)

Four of the 50 women with a preterm delivery had a spontaneous start of labor; the remaining had either induced labor (n=22) or a primary cesarean delivery (n=24), twenty-one being emergency cesarean deliveries (i.e. within 24 hours after making the decision). The median gestational age at delivery was 36^{+3} weeks in those who were induced and 31^{+3} weeks in those having a primary cesarean section. None of the fetuses were delivered preterm due to SGA alone, additional factors such as pre-eclampsia, abnormal CTG or fetal Doppler abnormalities were always present prior for induction or cesarean delivery. The frequency of extreme preterm delivery (<28 weeks) was 2.4% (n = 5), 15 (9.5%) fetuses were delivered between 28 and 33^{+6} weeks, and 30 were late preterm births (34 to 36^{+6} weeks).

In study II we used log-binomial regression analysis to test if size centiles and conditional growth centiles ≤5 and ≤10 were independently associated with adverse perinatal outcomes. Conditional growth centile ≤5 and ≤10 exerted independent effects on the following outcomes: preterm birth, operative delivery due to fetal distress, admission to the NICU, and the 'any adverse outcome' variable. Size centile ≤5 had independent association to operative delivery due to fetal distress and 'any adverse outcome' when adjusted for conditional growth centile. When cut off was set to 10^{th} centile, size was independently associated to admission to NICU as well. If the combination of size centiles and conditional growth centiles ≤5 and ≤10 significantly

improved prediction of adverse outcomes, compared to the use of SGA classification alone, were tested by log-likelihood test:

Adding conditional growth centile to size centile ≤5 in the model resulted in a significant improvement in the prediction of the following outcomes:

- preterm birth ($p = 0.023$)
- operative delivery due to fetal distress ($p = 0.028$)
- admission to the NICU ($p = 0.022$)
- 'any adverse outcome' ($p = 0.023$)

The combination of size centile and conditional growth centile ≤ 10 produced similar results in prediction of adverse outcomes:

- preterm birth ($p = 0.015$)
- operative delivery due to fetal distress ($p = 0.014$)
- admission to the NICU ($p = 0.024$)
- 'any adverse outcome' ($p = 0.012$)

Use of size centile ≤5 alone resulted in an identification of 66% of those with 'any adverse outcome'. Combining fetal size and conditional growth ≤5th centile resulted in an increase of 17% in the PPV; however, the improvement was not significantly increased. The sensitivity for fetal size centile ≤5 as predictor of 'any adverse outcome' was 60% and was not significantly changed when adding conditional growth centile in the model. While the specificity was significantly improved by combining size and conditional growth centile ≤5 compared to size centile alone (Table 5).

Table 5. Predictive values for 'any adverse outcome'.

	PPV % (95% Cl)	NPV % (95% Cl)	Sensitivity % (95% Cl)	Specificity % (95% Cl)
Size centile ≤ 5	66 (55 - 76)	73 (64 - 79)	60 (49 - 69)	78 (70 - 84)
Size and conditional growth centile ≤ 5	83 (69 - 92)	68 (61 - 75)	39 (30 - 50)	94 (89 - 97)

4.3 Study III

Serial measurements of MCA PI and CPR were obtained for 207 and 205 participants, respectively. Nineteen doctors with basic to advanced training in Doppler examination performed the measurements. A total of 865 observation of MCA PI was available and 851 observations of the CPR. The number of neonates with a birthweight ≤5th centile was 83; two of these had only one CPR observation and were not included in the analysis. The number of observations in these 83 SGA neonates was 311 for MCA PI and 304 for CPR. Of the SGA neonates 29% had an UA PI ≥95th centile, 31% had an MCA PI ≤5th centile and 47% had CPR centile ≤5 at the last visit. Distribution of AU and MCA PI at the final visit in SGA and AGA fetuses is presented in figure 5 and 6. In those with normal birthweight the frequency of pathologic Doppler was 4%, 6% and 7%, respectively.

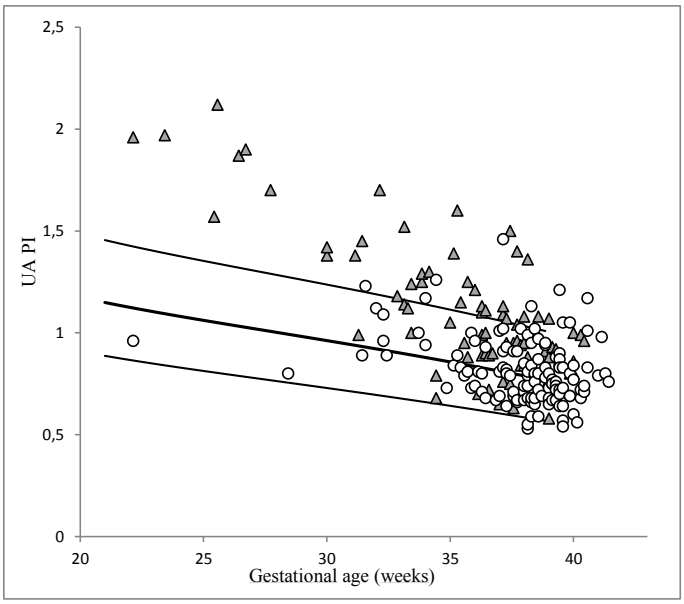


Figure 5. *Distribution of AU PI at the final visit in SGA fetuses (filled triangles) and in appropriate for gestational age (AGA) fetuses (open circles) in relation to reference curve for AU PI (139), the black lines representing the 95th, 50th and 5th centile.*

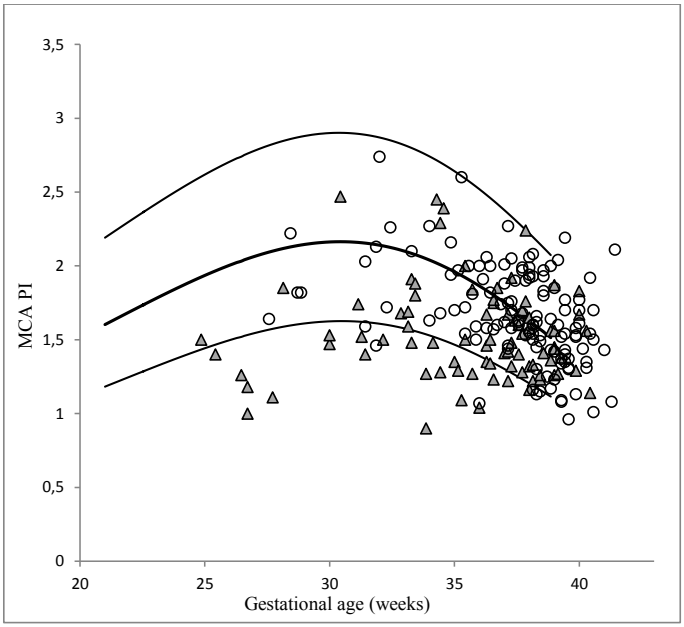


Figure 6. *Distribution of MCA PI at the final visit in SGA fetuses (filled triangles) and in appropriate for gestational age (AGA) fetuses (open circles) in relation to reference curve for MCA PI (140), the black lines representing the 95th, 50th and 5th centile.*

Maternal characteristics and birth outcomes are presented previously (same study population as for Study II).

In this study we found that conditional centiles ≤5 and ≤10 for MCA PI were associated with increased risk of preterm birth, operative delivery due to fetal distress, admission to NICU, 5 minute Apgar score <7 and the combined outcome variable 'any adverse outcome', results are presented in table 6.

Table 6. Log-binomial regression of conditional MCA PI centiles ≤5 and ≤10 and association with adverse outcomes in the total study (*n* = 207).

	Conditional centile ≤5 (*n* = 20)				Conditional centile ≤10 (*n* = 41)			
Outcomes	*n*	%	RR	95% CI	*n*	%	RR	95% CI
Preterm birth	16	80	4.5	3.1-6.6	27	66	5.0	3.2-7.8
OD due to fetal distress	14	70	3.6	2.4-5.5	24	59	3.7	2.4-5.8
NICU	15	75	4.4	2.9-6.6	27	66	5.5	3.4-8.7
5 min Apgar <7	3	15	4.0	1.1-14.3	6	15	6.1	1.8-20.5
'Any adverse outcome'	18	90	2.4	1.9-3.1*	32	78	2.4	1.8-3.1

* RR is calculated by cross table when regression analysis failed due to unsatisfied converge criteria.

OD, Operative delivery

When both conventional and conditional centiles were included in the model, conditional centiles for MCA PI ≤5 and ≤10 had no independent association with the outcomes. However, when including conditional centile ≤5 for MCA PI from the entire series of measurements in multilevel log-binomial regression analysis (639 pairs of observations) we demonstrated an independent effect and a doubled risk of operative delivery due to fetal distress, aRR 2.0 (95% CI; 1.1-3.4), but adding conditional centile ≤5 did not improve the prediction.

Similar to the analysis of MCA PI, the conditional centiles ≤5 and ≤10 for CPR was associated with adverse perinatal outcomes, except for low 5-min Apgar score (Table 7).

Table 7. Log-binomial regression analysis of conditional CPR centiles ≤5 and ≤10 and association with adverse outcomes in the total study population (*n* = 205).

	Conditional centile ≤5 (*n* = 38)				Conditional centile ≤10 (*n* = 51)			
Outcomes	*n*	%	RR	95% CI	*n*	%	RR	95% CI
Preterm birth	29	76	7.1	4.4-11.3	33	65	7.1	4.2-12.2
OD due to fetal distress	24	63	4.4	2.8-6.8	29	57	4.6	2.8-7.5
NICU	30	79	8.8	5.3-14.6	31	61	6.7	3.9-11.5
'Any adverse outcome'	34	90	2.9*	2.3-3.8	40	78	2.7	2.0-3.6

* RR is calculated by cross table when regression analysis failed due to unsatisfied converge criteria.

OD, Operative delivery

When both parameters were included in the model, the conventional and conditional centiles ≤10 had independent effects on risk of adverse perinatal outcomes, while conditional centile ≤5 for CPR had no independent effect on the outcomes. Combining conventional centile and conditional centile of CPR ≤10 resulted in a significant improvement (log-likelihood test) in the prediction of the following outcomes:

- Operative delivery due to fetal distress (p =0.032)
- Admission to NICU (p = 0.048)
- 'Any adverse outcome' (p = 0.034)

PPV for CPR ≤10th conventional centile in predicting 'any adverse outcome' was 73%, and an increase to 81% was seen when conditional centile ≤10 was added. However, the difference was not significant. The sensitivity and specificity for conventional centile of CPR ≤10 as predictor of 'any adverse outcome' were 52% and 87%,

respectively. Adding conditional centile of CPR ≤10 did not change sensitivity and specificity significantly (Table 8).

Table 8. Predictive ability of CPR ≤10th centile for 'any adverse outcome'.

	PPV % (95% Cl)	NPV % (95% Cl)	Sensitivity % (95% Cl)	Specificity % (95% Cl)
Conventional centile ≤10	73 (61 - 83)	72 (64 - 78)	52 (41 - 62)	87 (79 - 92)
Conventional and conditional centile ≤10	81 (68 - 90)	71 (63 - 77)	46 (36 - 56)	93 (86 - 96)

5 Discussion

5.1 Methodological consideration

5.1.1 Study design

Women in both study populations were included in prospective longitudinal studies. In study I information on maternal weight at the final visit were obtained from the antenatal chart and weight gain during pregnancy was then calculated. One single measurement of fetal AC in second trimester was used in the analysis. This prospective study design has the advantages of being standardized regarding variables like gestational age and measurement of the fetal AC, but the collected information on maternal weight is susceptible for biases. In study II and III the advantages of a prospective longitudinal design is exploited. Change in fetal size and circulation over a period of time expressed in conditional centiles was used to test the risk of adverse perinatal outcomes. Growth assessment and serial Doppler measurements requires reference ranges of a longitudinal design which were used in our studies. A prospective longitudinal study design is time consuming and has increased risk of withdrawal. This proved not to be a problem in our studies. Women in study II and III had high risk pregnancies, and would have been offered additional ultrasound examinations anyway. Most pregnant women appreciate additional ultrasound examinations during pregnancy; this can explain the low withdrawal rate in the low risk population of study I.

5.1.2 Biometry and Doppler measurements

In routine clinical practice a mean of three biometric measurements is used to reduce measurement error, this was also the case in all three studies. Inter -and intra-observer variation for the biometric measurements is published elsewhere (80). For Doppler measurements we used the measurements reported in the patient's records. Inter- and intra-observer variation for fetal Doppler measurements in the UA and MCA are presented in the publications of the references charts (139, 140). Inter –and intra-observer variation was not repeated in our studies (II and III), however there is reason

to believe that the inter-observer variation is somewhat higher when the number of doctors performing the examinations was relatively high.

Combs' formula for EFW was used to construct reference charts for fetal growth in the "Fetal age and growth" study (47), and this formula (52) is used in clinical practice in our department since it is evaluated to have good performance throughout pregnancy (256). At term an absolute error between EFW and birthweight of 7.6% is reported (256). The accuracy of birthweight prediction using EFW is reduced at low gestation and for both IUGR fetuses and in macrosomia (59, 257). The reference charts for EFW works when the intention is to identify fetuses that have a size outside the normal ranges. In such situations an ultrasound estimate of fetal size is compared to a reference chart based on intrauterine fetal size. If a reference chart based on birthweight is used, the 10^{th} centile will appear lower than the 10^{th} centile in a reference chart based on EFW due to increased frequency of pregnancy complications and IUGR in those who are born prematurely (105). As a consequence fewer growth restricted fetuses will be identified. In a recent French study, they have tested an alternative standard to better identify SGA fetuses at increased risk of perinatal mortality. They used a subpopulation-based birthweight standard where infants of mothers having conditions related to IUGR were excluded. Infants classified as SGA according to the subpopulation based birthweight standard, but which were not classified as SGA due to conventional birthweight standard, showed significantly increased risk of perinatal death. Those classified as SGA due to intrauterine reference curves but not by the subpopulation birthweight standard, showed no increase in perinatal death compared to normally sized neonates (106).

5.1.3 Internal validity

The causal relationship between an independent and a dependent variable will be dependent on the internal validity, which refers to systematic errors. There are several types of errors that can influence the internal validity such as selection bias, information bias and confounders.

Selection bias may occur if the effects on the dependent variable can be modified by differences between the study population and those not included in the study, but who were theoretically eligible for the study. In study I a highly selected low risk population was invited. Some selection bias is expected due to those who are willing to participate in such studies generally has better health than non-responders. Compared to data from 2002 registered in the MBR database mean maternal age were comparable (29.5 and 29.9 (MBR)) while the number of smokers was considerably lower in the study population, 8.6% compared to 22.0% in the general population. Information on maternal weight and height were not registered in the MFR database at the time the study was conducted. Gestational age at birth and birthweight were both somewhat higher in the study population, 282 vs 275 days and 3694 vs 3528 grams, respectively. These differences are expected when the intention is to create reference curves from a low risk population. For those with missing information on weight gain during pregnancy (n =101) other anthropometric data were comparable to the overall study population. In study II and III women who were referred due to increased risk of having an SGA fetus, or who were diagnosed with an SGA fetus, were invited to the study. However, all women at risk were not invited, which may be because they never were referred for additionally growth assessment or that the doctor examining them forgot to invite them. Among those without any known risk factor, which nevertheless had an SGA neonate, far from all were detected prenatally and were therefore not invited. There is a risk of selection bias due to increased referral and detection of the most severe cases of growth restriction. The outcomes are not expected to be shifted to a major extent when a large number of the participants had normally sized fetuses as well as fetuses with less severe growth restriction. However, severity of growth restriction influenced the frequency of examinations and could potentially have led to a shift in outcome variables such as preterm birth.

Information bias can occur when different sources or methods are used for data collection or variables are classified in a non-standardized way. In study I pre-pregnant weight was collected from the antenatal form, this information is prone to both information bias and recall bias. Standardized methods were used in the ultrasound

measurements in all three studies and clearly defined cut offs are used for the outcome variables in study II and III.

Confounding occurs when the observed association between the independent and the dependent variable actually is influenced by a third variable. In study I the positive association between maternal weight gain and fetal AC in second trimester may be influenced by differences in nutritional status and diet. Such information was not recorded during the study period and we had therefor no opportunity to adjust for such differences. Study II and III were clinical observational studies, and the managing clinicians were not blinded to ultrasound measurements of ethical reasons. In some cases sonographic findings led to increased examination frequency, which enhances the probability of earlier detection of abnormal CTG or Doppler findings, leading to earlier intervention and prematurity in some cases. Iatrogenic preterm birth further influenced other outcome variables such as admission to NICU. IUGR is closely related to hypertensive disorders such as HT and PE which was contributing indication in 25 (54%) of iatrogenic preterm deliveries. Some of these participants were delivered due to maternal indication alone, mainly due to severe PE. Maternal hypertensive disorders influenced the number of preterm birth in the group of fetuses >5th centile.

5.1.4 External validity

External validity refers to whether the findings can be generalized to other populations. In study I healthy pregnant women were included for a study that aimed to construct reference charts for fetal size and growth. There was no restriction regarding age, height, pre-pregnant weight or BMI and women who developed gestational complications such as diabetes, hypertension or PE were not excluded. Our findings can only be generalized to a low risk population of pregnant women. Participants in study II and III were included due to high risk of SGA or due to a finding of a fetus ≤5th centile and we believe the results are valid for other departments but restricted to high risk populations. The high number of doctors involved in the examinations and management increases the external validity.

5.2 Discussion of results

In study I we demonstrated a positive association between maternal weight gain during pregnancy and fetal AC in the second trimester in a low risk population. This early association suggests a common mechanism for maternal weight gain and fetal size. Maternal nutrition is known to influence pregnancy weight gain (258) in addition to impact the fetal liver circulation (90). Pregnant women with low body fat stores and those having an unhealthy diet (150) show an increased distribution of umbilical blood to the fetal liver, and less being shunted trough the DV. Low maternal weight gain during pregnancy influences liver blood flow distribution, the left liver lobe is prioritized on the expense of the right liver lobe (90). These findings are in line with our results. However, the association with fetal AC was only present in those with slow weight gain when the population was categorized in subgroups. Altered liver circulation will affect growth of the liver and production of IgF I and II. The area encompassed by the fetal AC is dominated by the fetal liver, suggesting that AC reflects the capacity of fetal liver metabolism. This may also be the reason why AC turned out to be the strongest predictor of adverse perinatal outcome in a recent study (211). It corroborates with the previous findings of changes in liver blood flow and associated fetal and neonatal body composition (259) and makes AC a promising surrogate for fetal metabolic status. There is growing evidence that fetal adaption to nutritional supply in pregnancy is linked to adult health (71, 231). The present study adds information that such a regulation can be traced as early as second trimester.

Fetal size and growth are obviously strongly related since size is a direct consequence of growth. Commonly, small fetal size or SGA is used to predict adverse outcomes. Depending on the cut off used, a certain proportion of these fetuses will be physiological small. Due to this, there is a need for methods that better identifies fetuses at risk. Intuitively fetal conditional growth centile will be a suitable parameter in predicting adverse outcomes, but existing literature does not unequivocally support its use. Therefore we have tested a prediction model for adverse perinatal outcomes using a combination of size and conditional growth centiles compared to the use of

SGA classification alone. Our results should encourage the use of conditional growth centiles in pregnancies at risk in clinical practice and research.

Although our study was not designed for testing the precision of predictors we calculated positive and negative predictive value, sensitivity and specificity for 'any adverse outcome'. Probability of 'any adverse outcome' was 83% when combining size and growth centiles ≤5 compared to 66% using size ≤5th centile alone. The difference of 17% was not significantly increased. As expected, sensitivity was reduced when adding conditional growth centile to size centile compared to the use of size centile alone. While sensitivity was not significantly changed, specificity improved from 78 to 94% by adding conditional growth centile to size centile. Identifying SGA fetuses at low risk is valuable in avoiding unnecessary surveillance and interventions.

Study III adds new information on serial observations of MCA PI and CPR, by applying conditional centiles, in prediction of adverse perinatal outcomes. The combination of conventional and conditional centile ≤10 for CPR significantly improved prediction of adverse perinatal outcomes compared to the use of conventional centile alone. Equally good results were not demonstrated for MCA PI, which is similar to existing literature (178, 179, 182), i.e. a better predictive value when using CPR. Our results support that low conditional centile for CPR is an independent predictor of adverse perinatal outcomes in addition to low conventional centile for CPR. However, further evidence of the usefulness of conditional centiles is needed to justify a recommendation for clinical practice. Should we suggest its practical application, it would be in cases with an indication for Doppler examination (e.g. fetal growth restriction). In such a case a CPR <5th centile is associated with increased perinatal risks. The addition of a conditional centile would reflect the development. Thus a normal conditional centile would be reassuring while ≤5 would indicate a deteriorating development. The system may be used in normally growing fetuses, but we expect the usefulness rather will be found in the high-risk group. Healthy fetuses operate in a wide range of normal cerebral blood flow (140), while

compromised fetuses are expected to narrow down variation during their response to compensate for the insufficient placental function. A next study with increased power is expected to give a clearer view of some of the effects that did not reach significance in the present study.

6 Conclusions

In study I we demonstrated a positive association between maternal weight gain in pregnancy and fetal AC in the second trimester in a low risk population, and the effect seemed to be strongest in mothers with the slowest weight gain.

In study II we showed that size centiles and conditional growth centiles ≤5 and ≤10 exert independent association with adverse neonatal outcomes such as preterm birth, operative delivery due to fetal distress and admission to NICU in a high-risk population. The prediction is improved by combining size and conditional growth centiles ≤5 and ≤10 compared with SGA classification alone. The study supports including conditional centiles of EFW in the clinical monitoring and management of IUGR.

In study III we have shown that conditional centile ≤10 of CPR has independent effect in prediction of adverse perinatal outcomes, and combining conventional and conditional centile improves prediction compared to the use of conventional centile alone. The results for MCA and CPR are less convincing than those for EFW, possibly due to the relatively high individual variation of these parameters also in the small fetuses, worth further exploration.

7 Future aspects

We have shown an association between maternal weight gain and fetal AC (which reflects fetal liver size) at the second trimester in a low risk population. This corroborates with the fact that fetal liver circulation is influenced by maternal diet, body composition and weight gain, indicating one possible mechanistic pathway that deserves further exploration. We are currently establishing a bio-bank (CONIMPREG) that includes nutritional information including biomarkers and registration of maternal activity. In this study, data collection starts before conception. We believe such information may give a more detailed knowledge of how growth trajectories for fetal growth are set and modified during the entire pregnancy.

Well-documented longitudinal reference ranges and models for calculating conditional centiles are available for both EFW and Doppler measurements. In a high-risk population we have shown significantly better prediction of adverse perinatal outcomes when adding conditional centiles to conventional centiles for EFW and CPR. Repeated measurements of fetal size are performed extensively, and our results support clinical use of conditional growth centiles in high risk pregnancies. Adverse perinatal outcomes do not occur exclusively in SGA fetuses or in pregnancies with predefined risk. Testing applicability of conditional growth centiles in a low risk-population would require a large number of participants. Sensitivity of prenatal identification of SGA fetuses in a low risk population is low with a detection rate varying between 20 and 57% (211). A combined evaluation of EFW and growth velocity of fetal AC has shown to better identify SGA fetuses with increased risk of neonatal morbidity (211). Further evaluation of conditional growth centiles for fetal AC in high risk pregnancies may be valuable for the identification of fetuses at risk of adverse perinatal outcomes.

During the present studies we have become increasingly aware of the effect of multiple serial measurements and of a possible reduction in the variance of MCA PI and CPR in extreme cases. We therefore plan a study of individual variation of cerebral blood flow in prediction of adverse perinatal outcomes, and a further study

exploiting the full set of serial measurements in sharpening individual prediction of outcome.

8 References

1. Brandt K. Lærebok i fødselshjælp: Aschehoug & Co; 1911.

2. Løvset JB, E. Lærebok i obstetrikk for jordmødre. Oslo: Aschehoug & Co; 1959.

3. Pedersen A. 100 år med redusert spedbarnsdødelighet https://www.ssb.no/helse/artikler-og-publikasjoner/100-aar-med-redusert-spedbarnsdodelighet: Statistisk sentralbyrå; 2003.

4. Roberts E, Wood P. Birth weight and adult health in historical perspective: evidence from a New Zealand cohort, 1907-1922. Social science & medicine. 2014;107:154-61.

5. Martinsen K. Omsorg, sykepleie og medisin. 2. ed: Universitetsforlaget; 2003.

6. Larssen K-EB, LS. Bergsjø, P. Finne, PH. Perinatal service in Norway during the 1970s. Trondheim: The Norwegian Institute for Hospital Research, 1981 NIS-Rapport 6/81.

7. Bergsjo P, Bakketeig LS, Langhoff-Roos J. The development of perinatal audit: 20 years' experience. Acta obstetricia et gynecologica Scandinavica. 2003;82(9):780-8.

8. Westin B. Gravidogram and fetal growth. Comparison with biochemical supervision. Acta obstetricia et gynecologica Scandinavica. 1977;56(4):273-82.

9. Wallin A, Gyllensward A, Westin B. Symphysis-fundus measurement in prediction of fetal growth disturbances. Acta obstetricia et gynecologica Scandinavica. 1981;60(3):317-23.

10. Rosenberg K, Grant JM, Tweedie I, Aitchison T, Gallagher F. Measurement of fundal height as a screening test for fetal growth retardation. British journal of obstetrics and gynaecology. 1982;89(6):447-50.

11. Bais JM, Eskes M, Pel M, Bonsel GJ, Bleker OP. Effectiveness of detection of intrauterine growth retardation by abdominal palpation as screening test in a low risk population: an observational study. European journal of obstetrics, gynecology, and reproductive biology. 2004;116(2):164-9.

12. Neilson JP. Symphysis-fundal height measurement in pregnancy. The Cochrane database of systematic reviews. 2000(2):CD000944.

13. Donald I, Macvicar J, Brown TG. Investigation of abdominal masses by pulsed ultrasound. Lancet. 1958;1(7032):1188-95.

14. Donald I, Brown TG. Demonstration of tissue interfaces within the body by ultrasonic echo sounding. The British journal of radiology. 1961;34:539-46.

15. Willocks J, Donald I, Campbell S, Dunsmore IR. Intrauterine growth assessed by ultrasonic foetal cephalometry. The Journal of obstetrics and gynaecology of the British Commonwealth. 1967;74(5):639-47.

16. Gulmezoglu AM, Crowther CA, Middleton P, Heatley E. Induction of labour for improving birth outcomes for women at or beyond term. The Cochrane database of systematic reviews. 2012;6:CD004945.

17. Morken NH, Klungsoyr K, Skjaerven R. Perinatal mortality by gestational week and size at birth in singleton pregnancies at and beyond term: a nationwide population-based cohort study. BMC pregnancy and childbirth. 2014;14:172.

18. Ingemarsson I, Kallen K. Stillbirths and rate of neonatal deaths in 76,761 postterm pregnancies in Sweden, 1982-1991: a register study. Acta obstetricia et gynecologica Scandinavica. 1997;76(7):658-62.

19. Wilcox AJ, Dunson D, Baird DD. The timing of the "fertile window" in the menstrual cycle: day specific estimates from a prospective study. Bmj. 2000;321(7271):1259-62.

20. Savitz DA, Terry JW, Jr., Dole N, Thorp JM, Jr., Siega-Riz AM, Herring AH. Comparison of pregnancy dating by last menstrual period, ultrasound scanning, and their combination. American journal of obstetrics and gynecology. 2002;187(6):1660-6.

21. Backe B. [Routine ultrasonography in obstetric care in Norway, 1994]. Tidsskrift for den Norske laegeforening : tidsskrift for praktisk medicin, ny raekke. 1997;117(16):2314-5.

22. Taipale P, Hiilesmaa V. Predicting delivery date by ultrasound and last menstrual period in early gestation. Obstetrics and gynecology. 2001;97(2):189-94.

23. Mongelli M, Wilcox M, Gardosi J. Estimating the date of confinement: ultrasonographic biometry versus certain menstrual dates. American journal of obstetrics and gynecology. 1996;174(1 Pt 1):278-81.

24. Bottomley C, Bourne T. Dating and growth in the first trimester. Best practice & research Clinical obstetrics & gynaecology. 2009;23(4):439-52.

25. Caughey AB, Nicholson JM, Washington AE. First- vs second-trimester ultrasound: the effect on pregnancy dating and perinatal outcomes. American journal of obstetrics and gynecology. 2008;198(6):703 e1-5; discussion e5-6.

26. Benson CB, Doubilet PM. Sonographic prediction of gestational age: accuracy of second- and third-trimester fetal measurements. AJR American journal of roentgenology. 1991;157(6):1275-7.

27. Abuhamad AZ, Bulletins-Obstetrics ACoP. ACOG Practice Bulletin, clinical management guidelines for obstetrician-gynecologists number 98, October 2008 (replaces Practice Bulletin number 58, December 2004). Ultrasonography in pregnancy. Obstetrics and gynecology. 2008;112(4):951-61.

28. Butt K, Lim K, Society of O, Gynaecologists of C. Determination of gestational age by ultrasound. Journal of obstetrics and gynaecology Canada : JOGC = Journal d'obstetrique et gynecologie du Canada : JOGC. 2014;36(2):171-83.

29. Robinson HP, Fleming JE. A critical evaluation of sonar "crown-rump length" measurements. British journal of obstetrics and gynaecology. 1975;82(9):702-10.

30. Verburg BO, Steegers EA, De Ridder M, Snijders RJ, Smith E, Hofman A, et al. New charts for ultrasound dating of pregnancy and assessment of fetal growth: longitudinal data from a population-based cohort study. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2008;31(4):388-96.

31. Hadlock FP, Shah YP, Kanon DJ, Lindsey JV. Fetal crown-rump length: reevaluation of relation to menstrual age (5-18 weeks) with high-resolution real-time US. Radiology. 1992;182(2):501-5.

32. Sladkevicius P, Saltvedt S, Almstrom H, Kublickas M, Grunewald C, Valentin L. Ultrasound dating at 12-14 weeks of gestation. A prospective cross-validation of established dating formulae in in-vitro fertilized pregnancies. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2005;26(5):504-11.

33. Committee on Obstetric Practice. Method for Estimating Due Date. American Institute of Ultrasound in Medicine. 2014.

34. WHO: recommended definitions, terminology and format for statistical tables related to the perinatal period and use of a new certificate for cause of perinatal deaths. Modifications recommended by FIGO as amended October 14, 1976. Acta obstetricia et gynecologica Scandinavica. 1977;56(3):247-53.

35. Bergsjo P, Denman DW, 3rd, Hoffman HJ, Meirik O. Duration of human singleton pregnancy. A population-based study. Acta obstetricia et gynecologica Scandinavica. 1990;69(3):197-207.

36. Kieler H, Axelsson O, Nilsson S, Waldenstrom U. The length of human pregnancy as calculated by ultrasonographic measurement of the fetal biparietal diameter. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1995;6(5):353-7.

37. Tunon K, Eik-Nes SH, Grottum P. A comparison between ultrasound and a reliable last menstrual period as predictors of the day of delivery in 15,000 examinations. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1996;8(3):178-85.

38. Johnsen SL, Wilsgaard T, Rasmussen S, Hanson MA, Godfrey KM, Kiserud T. Fetal size in the second trimester is associated with the duration of pregnancy, small fetuses having longer pregnancies. BMC pregnancy and childbirth. 2008;8:25.

39. Magnus P. Causes of variation in birth weight: a study of offspring of twins. Clinical genetics. 1984;25(1):15-24.

40. Little RE, Sing CF. Genetic and environmental influences on human birth weight. American journal of human genetics. 1987;40(6):512-26.

41. Godfrey K, Robinson S, Barker DJ, Osmond C, Cox V. Maternal nutrition in early and late pregnancy in relation to placental and fetal growth. Bmj. 1996;312(7028):410-4.

42. Simpson WJ. A preliminary report on cigarette smoking and the incidence of prematurity. American journal of obstetrics and gynecology. 1957;73(4):807-15.

43. Skjaerven R, Gjessing HK, Bakketeig LS. Birthweight by gestational age in Norway. Acta obstetricia et gynecologica Scandinavica. 2000;79(6):440-9.

44. Burton GJ, Hempstock J, Jauniaux E. Nutrition of the human fetus during the first trimester--a review. Placenta. 2001;22 Suppl A:S70-7.

45. Dickey RP, Gasser RF. Ultrasound evidence for variability in the size and development of normal human embryos before the tenth post-insemination week after assisted reproductive technologies. Human reproduction. 1993;8(2):331-7.

46. Smith GC, Smith MF, McNay MB, Fleming JE. First-trimester growth and the risk of low birth weight. The New England journal of medicine. 1998;339(25):1817-22.

47. Johnsen SL, Rasmussen S, Wilsgaard T, Sollien R, Kiserud T. Longitudinal reference ranges for estimated fetal weight. Acta obstetricia et gynecologica Scandinavica. 2006;85(3):286-97.

48. Pay A, Froen JF, Staff AC, Jacobsson B, Gjessing HK. Prediction of small-for-gestational-age status by symphysis-fundus height: a registry-based population cohort study. BJOG : an international journal of obstetrics and gynaecology. 2015.

49. Pay AS, Wiik J, Backe B, Jacobsson B, Strandell A, Klovning A. Symphysis-fundus height measurement to predict small-for-gestational-age status at birth: a systematic review. BMC pregnancy and childbirth. 2015;15:22.

50. Bergman E, Kieler H, Petzold MG, Sonesson C, Axelsson O. Symphysis-fundus measurements for detection of small for gestational age pregnancies. Acta obstetricia et gynecologica Scandinavica. 2006;85(4):407-12.

51. Backe B, Nakling J. Effectiveness of antenatal care: a population based study. British journal of obstetrics and gynaecology. 1993;100(8):727-32.

52. Combs CA, Jaekle RK, Rosenn B, Pope M, Miodovnik M, Siddiqi TA. Sonographic estimation of fetal weight based on a model of fetal volume. Obstetrics and gynecology. 1993;82(3):365-70.

53. Hadlock FP, Harrist RB, Sharman RS, Deter RL, Park SK. Estimation of fetal weight with the use of head, body, and femur measurements--a prospective study. American journal of obstetrics and gynecology. 1985;151(3):333-7.

54. Mielke G, Pietsch-Breitfeld B, Salinas R, Risse T, Marzusch K. A new formula for prenatal ultrasonographic weight estimation in extremely preterm fetuses. Gynecologic and obstetric investigation. 1995;40(2):84-8.

55. Dudley NJ, Lamb MP, Hatfield JA, Copping C, Sidebottom K. Estimated fetal weight in the detection of the small-for-menstrual-age fetus. Journal of clinical ultrasound : JCU. 1990;18(5):387-93.

56. Nahum GG, Stanislaw H. Ultrasonographic prediction of term birth weight: how accurate is it? American journal of obstetrics and gynecology. 2003;188(2):566-74.

57. Scioscia M, Vimercati A, Ceci O, Vicino M, Selvaggi LE. Estimation of birth weight by two-dimensional ultrasonography: a critical appraisal of its accuracy. Obstetrics and gynecology. 2008;111(1):57-65.

58. Ben-Haroush A, Yogev Y, Bar J, Mashiach R, Kaplan B, Hod M, et al. Accuracy of sonographically estimated fetal weight in 840 women with different pregnancy complications prior to induction of labor. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2004;23(2):172-6.

59. Abele H, Hoopmann M, Wagner N, Hahn M, Wallwiener D, Kagan KO. Accuracy of sonographic fetal weight estimation of fetuses with a birth weight of 1500 g or less. European journal of obstetrics, gynecology, and reproductive biology. 2010;153(2):131-7.

60. Dudley NJ. A systematic review of the ultrasound estimation of fetal weight. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2005;25(1):80-9.

61. Kritzer S, Magner K, Warshak CR. Increasing maternal body mass index and the accuracy of sonographic estimation of fetal weight near delivery. Journal of ultrasound in medicine : official journal of the American Institute of Ultrasound in Medicine. 2014;33(12):2173-9.

62. Farrell T, Holmes R, Stone P. The effect of body mass index on three methods of fetal weight estimation. BJOG : an international journal of obstetrics and gynaecology. 2002;109(6):651-7.

63. Barnhard Y, Bar-Hava I, Divon MY. Accuracy of intrapartum estimates of fetal weight. Effect of oligohydramnios. The Journal of reproductive medicine. 1996;41(12):907-10.

64. Benacerraf BR, Gelman R, Frigoletto FD, Jr. Sonographically estimated fetal weights: accuracy and limitation. American journal of obstetrics and gynecology. 1988;159(5):1118-21.

65. Chauhan SP, Scardo JA, Hendrix NW, Magann EF, Morrison JC. Accuracy of sonographically estimated fetal weight with and without oligohydramnios. A case-control study. The Journal of reproductive medicine. 1999;44(11):969-73.

66. Altman DG, Chitty LS. Charts of fetal size: 1. Methodology. British journal of obstetrics and gynaecology. 1994;101(1):29-34.

67. Royston P. Calculation of unconditional and conditional reference intervals for foetal size and growth from longitudinal measurements. Statistics in medicine. 1995;14(13):1417-36.

68. Royston P, Altman DG. Design and analysis of longitudinal studies of fetal size. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1995;6(5):307-12.

69. Owen P, Donnet ML, Ogston SA, Christie AD, Howie PW, Patel NB. Standards for ultrasound fetal growth velocity. British journal of obstetrics and gynaecology. 1996;103(1):60-9.

70. Kiserud T, Johnsen SL. Biometric assessment. Best practice & research Clinical obstetrics & gynaecology. 2009;23(6):819-31.

71. Gluckman PD, Hanson MA. Maternal constraint of fetal growth and its consequences. Seminars in fetal & neonatal medicine. 2004;9(5):419-25.

72. Clausson B, Lichtenstein P, Cnattingius S. Genetic influence on birthweight and gestational length determined by studies in offspring of twins. BJOG : an international journal of obstetrics and gynaecology. 2000;107(3):375-81.

73. Lunde A, Melve KK, Gjessing HK, Skjaerven R, Irgens LM. Genetic and environmental influences on birth weight, birth length, head circumference, and gestational age by use of population-based parent-offspring data. American journal of epidemiology. 2007;165(7):734-41.

74. Johnston LB, Clark AJ, Savage MO. Genetic factors contributing to birth weight. Archives of disease in childhood Fetal and neonatal edition. 2002;86(1):F2-3.

75. Gluckman PD, Pinal CS. Maternal-placental-fetal interactions in the endocrine regulation of fetal growth: role of somatotrophic axes. Endocrine. 2002;19(1):81-9.

76. Vasak B, Koenen SV, Koster MP, Hukkelhoven CW, Franx A, Hanson MA, et al. Human fetal growth is constrained below optimal for perinatal survival. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2015;45(2):162-7.

77. Morin I, Morin L, Zhang X, Platt RW, Blondel B, Breart G, et al. Determinants and consequences of discrepancies in menstrual and ultrasonographic gestational age estimates. BJOG : an international journal of obstetrics and gynaecology. 2005;112(2):145-52.

78. Sarris I, Bottomley C, Daemen A, Pexsters A, Timmerman D, Bourne T, et al. No influence of body mass index on first trimester fetal growth. Human reproduction. 2010;25(8):1895-9.

79. van Uitert EM, van der Elst-Otte N, Wilbers JJ, Exalto N, Willemsen SP, Eilers PH, et al. Periconception maternal characteristics and embryonic growth trajectories: the Rotterdam Predict study. Human reproduction. 2013;28(12):3188-96.

80. Johnsen SL, Wilsgaard T, Rasmussen S, Sollien R, Kiserud T. Longitudinal reference charts for growth of the fetal head, abdomen and femur. European journal of obstetrics, gynecology, and reproductive biology. 2006;127(2):172-85.

81. Ay L, Kruithof CJ, Bakker R, Steegers EA, Witteman JC, Moll HA, et al. Maternal anthropometrics are associated with fetal size in different periods of pregnancy and at birth. The Generation R Study. BJOG : an international journal of obstetrics and gynaecology. 2009;116(7):953-63.

82. Goldenberg RL, Davis RO, Cliver SP, Cutter GR, Hoffman HJ, Dubard MB, et al. Maternal risk factors and their influence on fetal anthropometric measurements. American journal of obstetrics and gynecology. 1993;168(4):1197-203; discussion 203-5.

83. In: Rasmussen KM, Yaktine AL, editors. Weight Gain During Pregnancy: Reexamining the Guidelines. The National Academies Collection: Reports funded by National Institutes of Health. Washington (DC)2009.

84. Seidman DS, Ever-Hadani P, Gale R. The effect of maternal weight gain in pregnancy on birth weight. Obstetrics and gynecology. 1989;74(2):240-6.

85. Sekiya N, Anai T, Matsubara M, Miyazaki F. Maternal weight gain rate in the second trimester are associated with birth weight and length of gestation. Gynecologic and obstetric investigation. 2007;63(1):45-8.

86. Abrams B, Selvin S. Maternal weight gain pattern and birth weight. Obstetrics and gynecology. 1995;86(2):163-9.

87. Durie DE, Thornburg LL, Glantz JC. Effect of second-trimester and third-trimester rate of gestational weight gain on maternal and neonatal outcomes. Obstetrics and gynecology. 2011;118(3):569-75.

88. Drehmer M, Duncan BB, Kac G, Schmidt MI. Association of second and third trimester weight gain in pregnancy with maternal and fetal outcomes. PloS one. 2013;8(1):e54704.

89. Rode L, Hegaard HK, Kjaergaard H, Moller LF, Tabor A, Ottesen B. Association between maternal weight gain and birth weight. Obstetrics and gynecology. 2007;109(6):1309-15.

90. Kessler J, Rasmussen S, Godfrey K, Hanson M, Kiserud T. Longitudinal study of umbilical and portal venous blood flow to the fetal liver: low pregnancy weight gain is associated with preferential supply to the fetal left liver lobe. Pediatric research. 2008;63(3):315-20.

91. Tchirikov M, Kertschanska S, Sturenberg HJ, Schroder HJ. Liver blood perfusion as a possible instrument for fetal growth regulation. Placenta. 2002;23 Suppl A:S153-8.

92. Ebbing C, Rasmussen S, Godfrey KM, Hanson MA, Kiserud T. Redistribution pattern of fetal liver circulation in intrauterine growth restriction. Acta obstetricia et gynecologica Scandinavica. 2009;88(10):1118-23.

93. Shu XO, Hatch MC, Mills J, Clemens J, Susser M. Maternal smoking, alcohol drinking, caffeine consumption, and fetal growth: results from a prospective study. Epidemiology. 1995;6(2):115-20.

94. Ellard GA, Johnstone FD, Prescott RJ, Ji-Xian W, Jian-Hua M. Smoking during pregnancy: the dose dependence of birthweight deficits. British journal of obstetrics and gynaecology. 1996;103(8):806-13.

95. Juarez SP, Merlo J. Revisiting the effect of maternal smoking during pregnancy on offspring birthweight: a quasi-experimental sibling analysis in Sweden. PloS one. 2013;8(4):e61734.

96. Little RE. Moderate alcohol use during pregnancy and decreased infant birth weight. American journal of public health. 1977;67(12):1154-6.

97. Cogswell ME, Yip R. The influence of fetal and maternal factors on the distribution of birthweight. Seminars in perinatology. 1995;19(3):222-40.

98. Wright JT, Waterson EJ, Barrison IG, Toplis PJ, Lewis IG, Gordon MG, et al. Alcohol consumption, pregnancy, and low birthweight. Lancet. 1983;1(8326 Pt 1):663-5.

99. Wiebe HW, Boule NG, Chari R, Davenport MH. The effect of supervised prenatal exercise on fetal growth: a meta-analysis. Obstetrics and gynecology. 2015;125(5):1185-94.

100. Kramer MS, Olivier M, McLean FH, Willis DM, Usher RH. Impact of intrauterine growth retardation and body proportionality on fetal and neonatal outcome. Pediatrics. 1990;86(5):707-13.

101. Bernstein IM, Horbar JD, Badger GJ, Ohlsson A, Golan A. Morbidity and mortality among very-low-birth-weight neonates with intrauterine growth restriction. The Vermont Oxford Network. American journal of obstetrics and gynecology. 2000;182(1 Pt 1):198-206.

102. McIntire DD, Bloom SL, Casey BM, Leveno KJ. Birth weight in relation to morbidity and mortality among newborn infants. The New England journal of medicine. 1999;340(16):1234-8.

103. Gluckman PD, Hanson MA. The consequences of being born small - an adaptive perspective. Hormone research. 2006;65 Suppl 3:5-14.

104. Ioannou C, Talbot K, Ohuma E, Sarris I, Villar J, Conde-Agudelo A, et al. Systematic review of methodology used in ultrasound studies aimed at creating charts of fetal size. BJOG : an international journal of obstetrics and gynaecology. 2012;119(12):1425-39.

105. Marsal K, Persson PH, Larsen T, Lilja H, Selbing A, Sultan B. Intrauterine growth curves based on ultrasonically estimated foetal weights. Acta paediatrica. 1996;85(7):843-8.

106. Ferdynus C, Quantin C, Abrahamowicz M, Burguet A, Sagot P, Gouyon JB. Comparison of the ability of alternative birthweight and fetal weight standards to identify preterm newborns at increased risk of perinatal death. BJOG : an international journal of obstetrics and gynaecology. 2013;120(12):1456-64.

107. Unterscheider J, Daly S, Geary MP, Kennelly MM, McAuliffe FM, O'Donoghue K, et al. Definition and management of fetal growth restriction: a survey of contemporary attitudes. European journal of obstetrics, gynecology, and reproductive biology. 2014;174:41-5.

108. Gynecologists RCoOa. The investigation and management of the small-for-gestational-age fetus (guideline no. 31). London: Royal College of Obstetricians and Gynecologists, 2014.

109. Unterscheider J, Daly S, Geary MP, Kennelly MM, McAuliffe FM, O'Donoghue K, et al. Optimizing the definition of intrauterine growth restriction: the multicenter prospective PORTO Study. American journal of obstetrics and gynecology. 2013;208(4):290 e1-6.

110. Campbell S, Thoms A. Ultrasound measurement of the fetal head to abdomen circumference ratio in the assessment of growth retardation. British journal of obstetrics and gynaecology. 1977;84(3):165-74.

111. Lin CC, Su SJ, River LP. Comparison of associated high-risk factors and perinatal outcome between symmetric and asymmetric fetal intrauterine growth retardation. American journal of obstetrics and gynecology. 1991;164(6 Pt 1):1535-41; discussion 41-2.

112. Dashe JS, McIntire DD, Lucas MJ, Leveno KJ. Effects of symmetric and asymmetric fetal growth on pregnancy outcomes. Obstetrics and gynecology. 2000;96(3):321-7.

113. Altman DG, Hytten FE. Intrauterine growth retardation: let's be clear about it. British journal of obstetrics and gynaecology. 1989;96(10):1127-32.

114. Long PA, Abell DA, Beischer NA. Fetal growth retardation and pre-eclampsia. British journal of obstetrics and gynaecology. 1980;87(1):13-8.

115. Odegard RA, Vatten LJ, Nilsen ST, Salvesen KA, Austgulen R. Preeclampsia and fetal growth. Obstetrics and gynecology. 2000;96(6):950-5.

116. Crovetto F, Crispi F, Scazzocchio E, Mercade I, Meler E, Figueras F, et al. First-trimester screening for early and late small-for-gestational-age neonates using maternal serum biochemistry, blood pressure and uterine artery Doppler. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2014;43(1):34-40.

117. Ebbing C, Kiserud T, Johnsen SL, Albrechtsen S, Rasmussen S. Prevalence, risk factors and outcomes of velamentous and marginal cord insertions: a population-based study of 634,741 pregnancies. PloS one. 2013;8(7):e70380.

118. Cooney MJ, Benson CB, Doubilet PM. Outcome of pregnancies in women with uterine duplication anomalies. Journal of clinical ultrasound : JCU. 1998;26(1):3-6.

119. Sibai BM, Abdella TN, Anderson GD. Pregnancy outcome in 211 patients with mild chronic hypertension. Obstetrics and gynecology. 1983;61(5):571-6.

120. Sibai BM, Lindheimer M, Hauth J, Caritis S, VanDorsten P, Klebanoff M, et al. Risk factors for preeclampsia, abruptio placentae, and adverse neonatal outcomes among women with chronic hypertension. National Institute of Child Health and Human Development Network of Maternal-Fetal Medicine Units. The New England journal of medicine. 1998;339(10):667-71.

121. McCowan LM, Buist RG, North RA, Gamble G. Perinatal morbidity in chronic hypertension. British journal of obstetrics and gynaecology. 1996;103(2):123-9.

122. Rey E, Couturier A. The prognosis of pregnancy in women with chronic hypertension. American journal of obstetrics and gynecology. 1994;171(2):410-6.

123. Cervera R, Font J, Carmona F, Balasch J. Pregnancy outcome in systemic lupus erythematosus: good news for the new millennium. Autoimmunity reviews. 2002;1(6):354-9.

124. Fink JC, Schwartz SM, Benedetti TJ, Stehman-Breen CO. Increased risk of adverse maternal and infant outcomes among women with renal disease. Paediatric and perinatal epidemiology. 1998;12(3):277-87.

125. Kimmerle R, Zass RP, Cupisti S, Somville T, Bender R, Pawlowski B, et al. Pregnancies in women with diabetic nephropathy: long-term outcome for mother and child. Diabetologia. 1995;38(2):227-35.

126. Haeri S, Khoury J, Kovilam O, Miodovnik M. The association of intrauterine growth abnormalities in women with type 1 diabetes mellitus complicated by vasculopathy. American journal of obstetrics and gynecology. 2008;199(3):278 e1-5.

127. Koubaa S, Hallstrom T, Lindholm C, Hirschberg AL. Pregnancy and neonatal outcomes in women with eating disorders. Obstetrics and gynecology. 2005;105(2):255-60.

128. Mills JL, Graubard BI, Harley EE, Rhoads GG, Berendes HW. Maternal alcohol consumption and birth weight. How much drinking during pregnancy is safe? Jama. 1984;252(14):1875-9.

129. Kirkinen P, Jouppila P, Herva R. Intra-uterine growth and fatal fetal abnormality. Acta obstetricia et gynecologica Scandinavica. 1983;62(1):43-7.

130. Snijders RJ, Sherrod C, Gosden CM, Nicolaides KH. Fetal growth retardation: associated malformations and chromosomal abnormalities. American journal of obstetrics and gynecology. 1993;168(2):547-55.

131. Levy RJ, Rosenthal A, Fyler DC, Nadas AS. Birthweight of infants with congenital heart disease. American journal of diseases of children. 1978;132(3):249-54.

132. Vandenbosche RC, Kirchner JT. Intrauterine growth retardation. American family physician. 1998;58(6):1384-90, 93-4.

133. Mongelli M, Ek S, Tambyrajia R. Screening for fetal growth restriction: a mathematical model of the effect of time interval and ultrasound error. Obstetrics and gynecology. 1998;92(6):908-12.

134. Wladimiroff JW, Tonge HM, Stewart PA. Doppler ultrasound assessment of cerebral blood flow in the human fetus. British journal of obstetrics and gynaecology. 1986;93(5):471-5.

135. Kiserud T, Acharya G. The fetal circulation. Prenatal diagnosis. 2004;24(13):1049-59.

136. Jensen A, Roman C, Rudolph AM. Effects of reducing uterine blood flow on fetal blood flow distribution and oxygen delivery. Journal of developmental physiology. 1991;15(6):309-23.

137. Yao AC, Moinian M, Lind J. Distribution of blood between infant and placenta after birth. Lancet. 1969;2(7626):871-3.

138. Gomez O, Figueras F, Fernandez S, Bennasar M, Martinez JM, Puerto B, et al. Reference ranges for uterine artery mean pulsatility index at 11-41 weeks of gestation. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2008;32(2):128-32.

139. Acharya G, Wilsgaard T, Berntsen GK, Maltau JM, Kiserud T. Reference ranges for serial measurements of umbilical artery Doppler indices in the second half of pregnancy. American journal of obstetrics and gynecology. 2005;192(3):937-44.

140. Ebbing C, Rasmussen S, Kiserud T. Middle cerebral artery blood flow velocities and pulsatility index and the cerebroplacental pulsatility ratio: longitudinal reference ranges and terms for serial measurements. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2007;30(3):287-96.

141. Kiserud T, Eik-Nes SH, Blaas HG, Hellevik LR. Ultrasonographic velocimetry of the fetal ductus venosus. Lancet. 1991;338(8780):1412-4.

142. Kessler J, Rasmussen S, Hanson M, Kiserud T. Longitudinal reference ranges for ductus venosus flow velocities and waveform indices. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2006;28(7):890-8.

143. Kiserud T, Eik-Nes SH, Blaas HG, Hellevik LR, Simensen B. Ductus venosus blood velocity and the umbilical circulation in the seriously growth-retarded fetus. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1994;4(2):109-14.

144. Hecher K, Campbell S, Doyle P, Harrington K, Nicolaides K. Assessment of fetal compromise by Doppler ultrasound investigation of the fetal circulation. Arterial, intracardiac, and venous blood flow velocity studies. Circulation. 1995;91(1):129-38.

145. Ferrazzi E, Bellotti M, Galan H, Pennati G, Bozzo M, Rigano S, et al. Doppler investigation in intrauterine growth restriction--from qualitative indices to flow measurements: a review of the experience of a collaborative group. Annals of the New York Academy of Sciences. 2001;943:316-25.

146. Oros D, Figueras F, Cruz-Martinez R, Meler E, Munmany M, Gratacos E. Longitudinal changes in uterine, umbilical and fetal cerebral Doppler indices in late-onset small-for-gestational age fetuses. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2011;37(2):191-5.

147. Baschat AA, Gembruch U, Harman CR. The sequence of changes in Doppler and biophysical parameters as severe fetal growth restriction worsens. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2001;18(6):571-7.

148. Figueras F, Gratacos E. Update on the diagnosis and classification of fetal growth restriction and proposal of a stage-based management protocol. Fetal diagnosis and therapy. 2014;36(2):86-98.

149. Flood K, Unterscheider J, Daly S, Geary MP, Kennelly MM, McAuliffe FM, et al. The role of brain sparing in the prediction of adverse outcomes in intrauterine growth restriction: results of the multicenter PORTO Study. American journal of obstetrics and gynecology. 2014;211(3):288 e1-5.

150. Haugen G, Hanson M, Kiserud T, Crozier S, Inskip H, Godfrey KM. Fetal liver-sparing cardiovascular adaptations linked to mother's slimness and diet. Circulation research. 2005;96(1):12-4.

151. Assali NS, Rauramo L, Peltonen T. Measurement of uterine blood flow and uterine metabolism. VIII. Uterine and fetal blood flow and oxygen consumption in early human pregnancy. American journal of obstetrics and gynecology. 1960;79:86-98.

152. Metcalfe J, Romney SL, Ramsey LH, Reid DE, Burwell CS. Estimation of uterine blood flow in normal human pregnancy at term. The Journal of clinical investigation. 1955;34(11):1632-8.

153. Pijnenborg R, Bland JM, Robertson WB, Brosens I. Uteroplacental arterial changes related to interstitial trophoblast migration in early human pregnancy. Placenta. 1983;4(4):397-413.

154. Pijnenborg R, Dixon G, Robertson WB, Brosens I. Trophoblastic invasion of human decidua from 8 to 18 weeks of pregnancy. Placenta. 1980;1(1):3-19.

155. Campbell S, Diaz-Recasens J, Griffin DR, Cohen-Overbeek TE, Pearce JM, Willson K, et al. New doppler technique for assessing uteroplacental blood flow. Lancet. 1983;1(8326 Pt 1):675-7.

156. Harrington K, Cooper D, Lees C, Hecher K, Campbell S. Doppler ultrasound of the uterine arteries: the importance of bilateral notching in the prediction of pre-eclampsia, placental abruption or delivery of a small-for-gestational-age baby. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1996;7(3):182-8.

157. Martin AM, Bindra R, Curcio P, Cicero S, Nicolaides KH. Screening for pre-eclampsia and fetal growth restriction by uterine artery Doppler at 11-14 weeks of gestation. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2001;18(6):583-6.

158. Albaiges G, Missfelder-Lobos H, Lees C, Parra M, Nicolaides KH. One-stage screening for pregnancy complications by color Doppler assessment of the uterine arteries at 23 weeks' gestation. Obstetrics and gynecology. 2000;96(4):559-64.

159. Papageorghiou AT, Yu CK, Bindra R, Pandis G, Nicolaides KH, Fetal Medicine Foundation Second Trimester Screening G. Multicenter screening for pre-eclampsia and fetal growth restriction by transvaginal uterine artery Doppler at 23 weeks of gestation. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2001;18(5):441-9.

160. Li N, Ghosh G, Gudmundsson S. Uterine artery Doppler in high-risk pregnancies at 23-24 gestational weeks is of value in predicting adverse outcome of pregnancy and selecting cases for more intense surveillance. Acta obstetricia et gynecologica Scandinavica. 2014;93(12):1276-81.

161. Gomez O, Martinez JM, Figueras F, Del Rio M, Borobio V, Puerto B, et al. Uterine artery Doppler at 11-14 weeks of gestation to screen for hypertensive disorders and associated complications in an unselected population. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2005;26(5):490-4.

162. Sutton MS, Gill T, Plappert T, Saltzman DH, Doubilet P. Assessment of right and left ventricular function in terms of force development with gestational age in the normal human fetus. British heart journal. 1991;66(4):285-9.

163. Kiserud T. Umbilical venous blood flow and reference ranges. Acta obstetricia et gynecologica Scandinavica. 2003;82(11):1061; author reply 2.

164. Su EJ. Role of the fetoplacental endothelium in fetal growth restriction with abnormal umbilical artery Doppler velocimetry. American journal of obstetrics and gynecology. 2015;213(4 Suppl):S123-30.

165. Trudinger BJ, Giles WB, Cook CM, Bombardieri J, Collins L. Fetal umbilical artery flow velocity waveforms and placental resistance: clinical significance. British journal of obstetrics and gynaecology. 1985;92(1):23-30.

166. Giles WB, Trudinger BJ, Baird PJ. Fetal umbilical artery flow velocity waveforms and placental resistance: pathological correlation. British journal of obstetrics and gynaecology. 1985;92(1):31-8.

167. Morrow RJ, Adamson SL, Bull SB, Ritchie JW. Effect of placental embolization on the umbilical arterial velocity waveform in fetal sheep. American journal of obstetrics and gynecology. 1989;161(4):1055-60.

168. Figueras F, Eixarch E, Meler E, Iraola A, Figueras J, Puerto B, et al. Small-for-gestational-age fetuses with normal umbilical artery Doppler have suboptimal perinatal and neurodevelopmental outcome. European journal of obstetrics, gynecology, and reproductive biology. 2008;136(1):34-8.

169. Severi FM, Bocchi C, Visentin A, Falco P, Cobellis L, Florio P, et al. Uterine and fetal cerebral Doppler predict the outcome of third-trimester small-for-gestational age fetuses with normal umbilical artery Doppler. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2002;19(3):225-8.

170. Kurkinen-Raty M, Kivela A, Jouppila P. The clinical significance of an absent end-diastolic velocity in the umbilical artery detected before the 34th week of pregnancy. Acta obstetricia et gynecologica Scandinavica. 1997;76(5):398-404.

171. Karsdorp VH, van Vugt JM, van Geijn HP, Kostense PJ, Arduini D, Montenegro N, et al. Clinical significance of absent or reversed end diastolic velocity waveforms in umbilical artery. Lancet. 1994;344(8938):1664-8.

172. Trudinger BJ, Cook CM, Giles WB, Ng S, Fong E, Connelly A, et al. Fetal umbilical artery velocity waveforms and subsequent neonatal outcome. British journal of obstetrics and gynaecology. 1991;98(4):378-84.

173. Morris RK, Malin G, Robson SC, Kleijnen J, Zamora J, Khan KS. Fetal umbilical artery Doppler to predict compromise of fetal/neonatal wellbeing in a high-risk population: systematic review and bivariate meta-analysis. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2011;37(2):135-42.

174. Wladimiroff JW, vd Wijngaard JA, Degani S, Noordam MJ, van Eyck J, Tonge HM. Cerebral and umbilical arterial blood flow velocity waveforms in normal and growth-retarded pregnancies. Obstetrics and gynecology. 1987;69(5):705-9.

175. Kiserud T. Physiology of the fetal circulation. Seminars in fetal & neonatal medicine. 2005;10(6):493-503.

176. Mari G, Deter RL. Middle cerebral artery flow velocity waveforms in normal and small-for-gestational-age fetuses. American journal of obstetrics and gynecology. 1992;166(4):1262-70.

177. Hershkovitz R, Kingdom JC, Geary M, Rodeck CH. Fetal cerebral blood flow redistribution in late gestation: identification of compromise in small fetuses with normal umbilical artery Doppler. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2000;15(3):209-12.

178. Gramellini D, Folli MC, Raboni S, Vadora E, Merialdi A. Cerebral-umbilical Doppler ratio as a predictor of adverse perinatal outcome. Obstetrics and gynecology. 1992;79(3):416-20.

179. Cruz-Martinez R, Figueras F, Hernandez-Andrade E, Oros D, Gratacos E. Fetal brain Doppler to predict cesarean delivery for nonreassuring fetal status in term small-for-gestational-age fetuses. Obstetrics and gynecology. 2011;117(3):618-26.

180. Khalil A, Morales-Rosello J, Townsend R, Morlando M, Papageorghiou A, Bhide A, et al. Value of third-trimester cerebroplacental ratio and uterine artery Doppler indices as predictors of stillbirth and perinatal loss. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2016;47(1):74-80.

181. Bahado-Singh RO, Kovanci E, Jeffres A, Oz U, Deren O, Copel J, et al. The Doppler cerebroplacental ratio and perinatal outcome in intrauterine growth restriction. American journal of obstetrics and gynecology. 1999;180(3 Pt 1):750-6.

182. Odibo AO, Riddick C, Pare E, Stamilio DM, Macones GA. Cerebroplacental Doppler ratio and adverse perinatal outcomes in intrauterine growth restriction: evaluating the impact of using gestational age-specific reference values. Journal of ultrasound in medicine : official journal of the American Institute of Ultrasound in Medicine. 2005;24(9):1223-8.

183. Morales-Rosello J, Khalil A, Morlando M, Hervas-Marin D, Perales-Marin A. Doppler reference values of the fetal vertebral and middle cerebral arteries, at 19-41 weeks gestation. The journal of maternal-fetal & neonatal medicine : the official journal of the European Association of Perinatal Medicine, the Federation of Asia and Oceania Perinatal Societies, the International Society of Perinatal Obstet. 2015;28(3):338-43.

184. Karlsen HO, Johnsen SL, Rasmussen S, Kiserud T. Prediction of adverse neonatal outcomes using size centiles and conditional growth centiles. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2015.

185. Kiserud T, Rasmussen S, Skulstad S. Blood flow and the degree of shunting through the ductus venosus in the human fetus. American journal of obstetrics and gynecology. 2000;182(1 Pt 1):147-53.

186. Kiserud T, Kessler J, Ebbing C, Rasmussen S. Ductus venosus shunting in growth-restricted fetuses and the effect of umbilical circulatory compromise. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2006;28(2):143-9.

187. Kessler J, Rasmussen S, Godfrey K, Hanson M, Kiserud T. Fetal growth restriction is associated with prioritization of umbilical blood flow to the left hepatic lobe at the expense of the right lobe. Pediatric research. 2009;66(1):113-7.

188. Hecher K, Snijders R, Campbell S, Nicolaides K. Fetal venous, intracardiac, and arterial blood flow measurements in intrauterine growth retardation: relationship with fetal blood gases. American journal of obstetrics and gynecology. 1995;173(1):10-5.

189. Baschat AA, Cosmi E, Bilardo CM, Wolf H, Berg C, Rigano S, et al. Predictors of neonatal outcome in early-onset placental dysfunction. Obstetrics and gynecology. 2007;109(2 Pt 1):253-61.

190. Hecher K, Bilardo CM, Stigter RH, Ville Y, Hackeloer BJ, Kok HJ, et al. Monitoring of fetuses with intrauterine growth restriction: a longitudinal study. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2001;18(6):564-70.

191. Lees C, Marlow N, Arabin B, Bilardo CM, Brezinka C, Derks JB, et al. Perinatal morbidity and mortality in early-onset fetal growth restriction: cohort outcomes of the trial of randomized umbilical and fetal flow in Europe (TRUFFLE). Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2013;42(4):400-8.

192. Tchirikov M, Kertschanska S, Schroder HJ. Obstruction of ductus venosus stimulates cell proliferation in organs of fetal sheep. Placenta. 2001;22(1):24-31.

193. Ong K, Kratzsch J, Kiess W, Costello M, Scott C, Dunger D. Size at birth and cord blood levels of insulin, insulin-like growth factor I (IGF-I), IGF-II, IGF-binding protein-1 (IGFBP-1), IGFBP-3, and the soluble IGF-II/mannose-6-phosphate receptor in term human infants. The ALSPAC Study Team. Avon Longitudinal Study of Pregnancy and Childhood. The Journal of clinical endocrinology and metabolism. 2000;85(11):4266-9.

194. Barker DJ. Intrauterine programming of adult disease. Molecular medicine today. 1995;1(9):418-23.

195. Baschat AA. Fetal growth restriction - from observation to intervention. Journal of perinatal medicine. 2010;38(3):239-46.

196. Group GS. A randomised trial of timed delivery for the compromised preterm fetus: short term outcomes and Bayesian interpretation. BJOG : an international journal of obstetrics and gynaecology. 2003;110(1):27-32.

197. Boers KE, Vijgen SM, Bijlenga D, van der Post JA, Bekedam DJ, Kwee A, et al. Induction versus expectant monitoring for intrauterine growth restriction at term: randomised equivalence trial (DIGITAT). Bmj. 2010;341:c7087.

198. Fu J, Olofsson P. Restrained cerebral hyperperfusion in response to superimposed acute hypoxemia in growth-restricted human fetuses with established brain-sparing blood flow. Early human development. 2006;82(3):211-6.

199. Baschat AA, Gembruch U, Reiss I, Gortner L, Weiner CP, Harman CR. Relationship between arterial and venous Doppler and perinatal outcome in fetal growth restriction. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2000;16(5):407-13.

200. Yli BM, Kessler J, Eikeland T, Hustad BL, Dragnes W, Henriksen T. What is the gold standard for intrapartum fetal monitoring? Acta obstetricia et gynecologica Scandinavica. 2012;91(9):1011-4.

201. Amer-Wahlin I, Hellsten C, Noren H, Hagberg H, Herbst A, Kjellmer I, et al. Cardiotocography only versus cardiotocography plus ST analysis of fetal electrocardiogram for intrapartum fetal monitoring: a Swedish randomised controlled trial. Lancet. 2001;358(9281):534-8.

202. Prior T, Mullins E, Bennett P, Kumar S. Prediction of intrapartum fetal compromise using the cerebroumbilical ratio: a prospective observational study. American journal of obstetrics and gynecology. 2013;208(2):124 e1-6.

203. Owen P, Harrold AJ, Farrell T. Fetal size and growth velocity in the prediction of intrapartum caesarean section for fetal distress. British journal of obstetrics and gynaecology. 1997;104(4):445-9.

204. Simchen MJ, Beiner ME, Strauss-Liviathan N, Dulitzky M, Kuint J, Mashiach S, et al. Neonatal outcome in growth-restricted versus appropriately grown preterm infants. American journal of perinatology. 2000;17(4):187-92.

205. Regev RH, Lusky A, Dolfin T, Litmanovitz I, Arnon S, Reichman B, et al. Excess mortality and morbidity among small-for-gestational-age premature infants: a population-based study. The Journal of pediatrics. 2003;143(2):186-91.

206. Gluckman PD, Hanson MA. The developmental origins of the metabolic syndrome. Trends in endocrinology and metabolism: TEM. 2004;15(4):183-7.

207. de Waal CG, Weisglas-Kuperus N, van Goudoever JB, Walther FJ, NeoNed Study G, Group LNFS. Mortality, neonatal morbidity and two year follow-up of extremely preterm infants born in The Netherlands in 2007. PloS one. 2012;7(7):e41302.

208. Vergani P, Roncaglia N, Ghidini A, Crippa I, Cameroni I, Orsenigo F, et al. Can adverse neonatal outcome be predicted in late preterm or term fetal growth restriction? Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2010;36(2):166-70.

209. Pilliod RA, Cheng YW, Snowden JM, Doss AE, Caughey AB. The risk of intrauterine fetal death in the small-for-gestational-age fetus. American journal of obstetrics and gynecology. 2012;207(4):318 e1-6.

210. Froen JF, Gardosi JO, Thurmann A, Francis A, Stray-Pedersen B. Restricted fetal growth in sudden intrauterine unexplained death. Acta obstetricia et gynecologica Scandinavica. 2004;83(9):801-7.

211. Sovio U, White IR, Dacey A, Pasupathy D, Smith GC. Screening for fetal growth restriction with universal third trimester ultrasonography in nulliparous women in the Pregnancy Outcome Prediction (POP) study: a prospective cohort study. Lancet. 2015.

212. Mihu D, Diculescu D, Costin N, Mihu CM, Blaga L, Ciortea R, et al. Applications of Doppler ultrasound during labor. Medical ultrasonography. 2011;13(2):141-9.

213. Aldrich CJ, D'Antona D, Spencer JA, Wyatt JS, Peebles DM, Delpy DT, et al. The effect of maternal pushing on fetal cerebral oxygenation and blood volume during the second stage of labour. British journal of obstetrics and gynaecology. 1995;102(6):448-53.

214. Schneider H, Progler M, Ziegler WH, Huch R. Biochemical changes in the mother and the fetus during labor and its significance for the management of the second stage. International journal of gynaecology and obstetrics: the official organ of the International Federation of Gynaecology and Obstetrics. 1990;31(2):117-26.

215. Thorngren-Jerneck K, Herbst A. Low 5-minute Apgar score: a population-based register study of 1 million term births. Obstetrics and gynecology. 2001;98(1):65-70.

216. Lie KK, Groholt EK, Eskild A. Association of cerebral palsy with Apgar score in low and normal birthweight infants: population based cohort study. Bmj. 2010;341:c4990.

217. Low JA, Panagiotopoulos C, Derrick EJ. Newborn complications after intrapartum asphyxia with metabolic acidosis in the term fetus. American journal of obstetrics and gynecology. 1994;170(4):1081-7.

218. Low JA. Intrapartum fetal asphyxia: definition, diagnosis, and classification. American journal of obstetrics and gynecology. 1997;176(5):957-9.

219. Blair E, Stanley FJ. Intrapartum asphyxia: a rare cause of cerebral palsy. The Journal of pediatrics. 1988;112(4):515-9.

220. Hafstrom M, Ehnberg S, Blad S, Noren H, Renman C, Rosen KG, et al. Developmental outcome at 6.5 years after acidosis in term newborns: a population-based study. Pediatrics. 2012;129(6):e1501-7.

221. Mejri A, Dorval VG, Nuyt AM, Carceller A. Hypoglycemia in term newborns with a birth weight below the 10th percentile. Paediatrics & child health. 2010;15(5):271-5.

222. Rozance PJ, Hay WW. Hypoglycemia in newborn infants: Features associated with adverse outcomes. Biology of the neonate. 2006;90(2):74-86.

223. Mansson J, Stjernqvist K. Children born extremely preterm show significant lower cognitive, language and motor function levels compared with children born at term, as measured by the Bayley-III at 2.5 years. Acta paediatrica. 2014;103(5):504-11.

224. Baschat AA. Neurodevelopment following fetal growth restriction and its relationship with antepartum parameters of placental dysfunction. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2011;37(5):501-14.

225. Brodszki J, Morsing E, Malcus P, Thuring A, Ley D, Marsal K. Early intervention in management of very preterm growth-restricted fetuses: 2-year outcome of infants delivered on fetal indication before 30 gestational weeks. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2009;34(3):288-96.

226. Leitner Y, Fattal-Valevski A, Geva R, Bassan H, Posner E, Kutai M, et al. Six-year follow-up of children with intrauterine growth retardation: long-term, prospective study. Journal of child neurology. 2000;15(12):781-6.

227. Leitner Y, Fattal-Valevski A, Geva R, Eshel R, Toledano-Alhadef H, Rotstein M, et al. Neurodevelopmental outcome of children with intrauterine growth retardation: a longitudinal, 10-year prospective study. Journal of child neurology. 2007;22(5):580-7.

228. Arcangeli T, Thilaganathan B, Hooper R, Khan KS, Bhide A. Neurodevelopmental delay in small babies at term: a systematic review. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 2012;40(3):267-75.

229. Lees CC, Marlow N, van Wassenaer-Leemhuis A, Arabin B, Bilardo CM, Brezinka C, et al. 2 year neurodevelopmental and intermediate perinatal outcomes in infants with very preterm fetal growth restriction (TRUFFLE): a randomised trial. Lancet. 2015;385(9983):2162-72.

230. Thornton JG, Hornbuckle J, Vail A, Spiegelhalter DJ, Levene M, group Gs. Infant wellbeing at 2 years of age in the Growth Restriction Intervention Trial (GRIT): multicentred randomised controlled trial. Lancet. 2004;364(9433):513-20.

231. Barker DJ, Gluckman PD, Godfrey KM, Harding JE, Owens JA, Robinson JS. Fetal nutrition and cardiovascular disease in adult life. Lancet. 1993;341(8850):938-41.

232. Gluckman PD, Cutfield W, Hofman P, Hanson MA. The fetal, neonatal, and infant environments-the long-term consequences for disease risk. Early human development. 2005;81(1):51-9.

233. Gluckman PD, Hanson MA, Pinal C. The developmental origins of adult disease. Maternal & child nutrition. 2005;1(3):130-41.

234. Leon DA, Lithell HO, Vagero D, Koupilova I, Mohsen R, Berglund L, et al. Reduced fetal growth rate and increased risk of death from ischaemic heart disease: cohort study of 15 000 Swedish men and women born 1915-29. Bmj. 1998;317(7153):241-5.

235. Huxley RR, Shiell AW, Law CM. The role of size at birth and postnatal catch-up growth in determining systolic blood pressure: a systematic review of the literature. Journal of hypertension. 2000;18(7):815-31.

236. Ramadhani MK, Grobbee DE, Bots ML, Castro Cabezas M, Vos LE, Oren A, et al. Lower birth weight predicts metabolic syndrome in young adults: the Atherosclerosis Risk in Young Adults (ARYA)-study. Atherosclerosis. 2006;184(1):21-7.

237. Hovi P, Andersson S, Jarvenpaa AL, Eriksson JG, Strang-Karlsson S, Kajantie E, et al. Decreased bone mineral density in adults born with very low birth weight: a cohort study. PLoS medicine. 2009;6(8):e1000135.

238. Cooper C, Javaid MK, Taylor P, Walker-Bone K, Dennison E, Arden N. The fetal origins of osteoporotic fracture. Calcified tissue international. 2002;70(5):391-4.

239. Whitworth M, Bricker L, Neilson JP, Dowswell T. Ultrasound for fetal assessment in early pregnancy. The Cochrane database of systematic reviews. 2010(4):CD007058.

240. van Dyk B, Motto JA, Buchmann EJ. Routine second-trimester ultrasound for low risk pregnancies in a South African community. International journal of gynaecology and obstetrics: the official organ of the International Federation of Gynaecology and Obstetrics. 2007;98(3):257-8.

241. Salvesen KA, Vatten LJ, Jacobsen G, Eik-Nes SH, Okland O, Molne K, et al. Routine ultrasonography in utero and subsequent vision and hearing at primary school age. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1992;2(4):243-4, 5-7.

242. Salvesen KA, Bakketeig LS, Eik-nes SH, Undheim JO, Okland O. Routine ultrasonography in utero and school performance at age 8-9 years. Lancet. 1992;339(8785):85-9.

243. Salvesen KA, Vatten LJ, Bakketeig LS, Eik-Nes SH. Routine ultrasonography in utero and speech development. Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology. 1994;4(2):101-3.

244. Kieler H, Cnattingius S, Haglund B, Palmgren J, Axelsson O. Sinistrality--a side-effect of prenatal sonography: a comparative study of young men. Epidemiology. 2001;12(6):618-23.

245. Stark CR, Orleans M, Haverkamp AD, Murphy J. Short- and long-term risks after exposure to diagnostic ultrasound in utero. Obstetrics and gynecology. 1984;63(2):194-200.

246. Salvesen KA. Epidemiological prenatal ultrasound studies. Progress in biophysics and molecular biology. 2007;93(1-3):295-300.

247. Stalberg K, Haglund B, Axelsson O, Cnattingius S, Pfeifer S, Kieler H. Prenatal ultrasound and the risk of childhood brain tumour and its subtypes. British journal of cancer. 2008;98(7):1285-7.

248. Salvesen KA, Vatten LJ, Eik-Nes SH, Hugdahl K, Bakketeig LS. Routine ultrasonography in utero and subsequent handedness and neurological development. Bmj. 1993;307(6897):159-64.

249. Newnham JP, Evans SF, Michael CA, Stanley FJ, Landau LI. Effects of frequent ultrasound during pregnancy: a randomised controlled trial. Lancet. 1993;342(8876):887-91.

250. Sande RK, Matre K, Eide GE, Kiserud T. The effect of ultrasound output level on obstetric biometric measurements. Ultrasound in medicine & biology. 2013;39(1):37-43.

251. Sande RK, Matre K, Eide GE, Kiserud T. The effects of reducing the thermal index for bone from 1.0 to 0.5 and 0.1 on common obstetric pulsed wave Doppler measurements in the second half of pregnancy. Acta obstetricia et gynecologica Scandinavica. 2013;92(7):790-6.

252. Johnsen SL, Rasmussen S, Sollien R, Kiserud T. Fetal age assessment based on ultrasound head biometry and the effect of maternal and fetal factors. Acta obstetricia et gynecologica Scandinavica. 2004;83(8):716-23.

253. Altman DG. Practical Statistics for Medical Research. London: Chapman & Hall; 1999.

254. Voskamp BJ, Kazemier BM, Ravelli AC, Schaaf J, Mol BW, Pajkrt E. Recurrence of small-for-gestational-age pregnancy: analysis of first and subsequent singleton pregnancies in The Netherlands. American journal of obstetrics and gynecology. 2013;208(5):374 e1-6.

255. Williams R. Multicollinearity [Internet]. http://www3.nd.edu/~rwilliam/ 2015.

256. Mongelli M, Biswas A. Menstrual age-dependent systematic error in sonographic fetal weight estimation: a mathematical model. Journal of clinical ultrasound : JCU. 2002;30(3):139-44.

257. Alsulyman OM, Ouzounian JG, Kjos SL. The accuracy of intrapartum ultrasonographic fetal weight estimation in diabetic pregnancies. American journal of obstetrics and gynecology. 1997;177(3):503-6.

258. Thangaratinam S, Rogozinska E, Jolly K, Glinkowski S, Roseboom T, Tomlinson JW, et al. Effects of interventions in pregnancy on maternal weight and obstetric outcomes: meta-analysis of randomised evidence. Bmj. 2012;344:e2088.

259. Godfrey KM, Haugen G, Kiserud T, Inskip HM, Cooper C, Harvey NC, et al. Fetal liver blood flow distribution: role in human developmental strategy to prioritize fat deposition versus brain development. PloS one. 2012;7(8):e41759.

I

AOGS
ACTA Obstetricia et Gynecologica Scandinavica

MAIN RESEARCH ARTICLE

Maternal weight gain: a determinant for fetal abdominal circumference in the second trimester

HENRIETTE HELLEBUST[1], SYNNØVE LIAN JOHNSEN[1], SVEIN RASMUSSEN[1,2] & TORVID KISERUD[1,2]

[1]Department of Obstetrics and Gynaecology, Haukeland University Hospital, Bergen, [2]Department of Clinical Medicine, University of Bergen, Norway

Key words
Fetal abdominal circumference, growth, maternal weight gain, nutrition, ultrasound biometry

Correspondence
Dr. Henriette Hellebust, Department of Obstetrics and Gynecology, Haukeland University Hospital, Jonas Lies Vei 65, Bergen 5021, Norway. E-mail: henriette.hellebust@helse-bergen.no

Conflict of interest
The authors have stated explicitly that there are no conflicts of interest in connection with this article and that they are solely responsible for the content and writing of this paper.

Received: 18 November 2010
Accepted: 11 March 2011

DOI: 10.1111/j.1600-0412.2011.01129.x

Abstract

Objective. To study the association between maternal weight gain in pregnancy and fetal abdominal circumference in the second trimester. *Design.* Prospective cross-sectional study. *Setting.* Low-risk antenatal clinic. *Population.* Six hundred and fifty women with low-risk pregnancy. *Methods.* Women with a regular menstrual period (28±4 days) and certain information on the last menstrual period were recruited when they were referred for routine ultrasound scanning. Women with a discrepancy of>14 days between ultrasound and menstrual age were excluded. Maternal weight gain during pregnancy was derived from information in the antenatal chart and the weekly weight gain was calculated. Fetal abdominal circumference measurements were registered in gestational weeks 15–25 and their z-scores, together with the z-scores of maternal weight gain, were used in a linear regression analysis. *Main outcome measures.* Association between maternal weight gain and fetal abdominal circumference. *Results.* Based on the complete data of 515 women we found a mean maternal weight gain during pregnancy of 0.39kg/week and a positive association between this weight gain and fetal abdominal circumference in the second trimester (r=0.122 (95%CI 0.051–0.194)), with the strongest effect in women with the slowest weight gain (<0.28kg/week) (r=0.554 (95%CI 0.261–0.846)). *Conclusion.* Maternal weight gain in pregnancy is related to and may determine fetal abdominal circumference in gestational weeks 15–25, particularly in those women with a slow weight gain.

Abbreviations: AC, abdominal circumference; BMI, body mass index; LMP, last menstrual period; CRL, crown rump length; BPD, biparietal diameter.

Introduction

Birthweight shows considerable variation and has in recent years been linked to various health risks in adult life (1). This has led to an increased interest in identifying mechanisms and factors that determine fetal growth. Maternal constraint on fetal growth has been shown to be the most prominent factor (2). Maternal height, body mass index (BMI) and weight gain during pregnancy are shown to be part of this by influencing estimated fetal weight and weight at birth (3–5).

The mechanism through which these factors are operational is not known, but recent data suggest that the fetal liver is a key organ translating maternal factors into differential growth (6). Umbilical liver perfusion, liver proliferation and production of insulin-like growth factors are important components of this mechanism (7–9). Measuring the fetal liver has been attempted and a relation between liver volume and fetal growth has been established (10) but the technique is time-consuming and is hampered by high variability. However, as the fetal liver is the dominating organ included in the abdominal circumference measurement (AC), the AC could be used to express liver size. AC additionally includes subcutaneous and intra-abdominal fat accretion, another expression of growth variation, and another reason why we used AC in the present study to test our hypothesis that maternal weight gain influences AC in the second trimester.

Among the maternal factors, the maternal gestational weight gain is of particular interest. It reflects growth of the conceptus (including the fetus) and maternal tissue expansion but also nutrition during pregnancy. The appropriate

weight gain is regarded to be 11.5–16kg for women with a normal BMI (11), and it is well established that there is a positive relation between total weight gain in pregnancy and birthweight (12–14). However, only one study has explored the relation between maternal weight gain in pregnancy and AC. That study showed that low total weight gain (<8kg) compared with a high (>16kg) weight gain, was associated with a difference in fetal AC of just 1.4%, but no effect was seen at 18 weeks' gestation (4).

Thus, the aim of the present study was to assess whether maternal weight gain during pregnancy might influence fetal AC as early as in the second trimester in low risk pregnancies.

Material and methods

The present study is part of the 'Fetal Age and Growth' project that included 650 women with low-risk pregnancies for the ultrasound study of fetal size, pregnancy duration and fetal growth published previously (5,15–17). Here we used the information on maternal weight gain not published previously to address our hypothesis of a nutritional influence on the fetal AC as a surrogate for liver size and fat accretion in the second trimester. The Regional Committee of Medical Research Ethics approved the study (REK-III no. 025.01). The women participated voluntarily and gave their written informed consent at entrance. Inclusion criteria were: regular menstrual periods (28±4 days) for at least three months before pregnancy and no use of hormone therapy in this period, an exact known date of the last menstrual period (LMP), singleton pregnancy, no history of complications in previous pregnancies, no regular use of medication. The exclusion criterion was>14 days' discrepancy between ultrasound and menstrual age.

Information on maternal pre-pregnancy weight had been recorded at the first antenatal visit. Weight gain during pregnancy was calculated as the difference between weight at the last antenatal visit and the pre-pregnancy weight. For the analysis we calculated weight gain per week until the last antenatal visit. Gestational age was assessed using the LMP.

All ultrasound examinations were performed by two experienced persons, using a Philips HDI 5000 machine (Philips, Seattle, WA, USA), with a 2–5MHz abdominal scanning head, or an Aloka Prosound-5000 machine (Aloka, Tokyo, Japan), with a 2–5MHz abdominal scanning head.

Fetal AC ultrasound measurements were obtained using an ellipse in the transverse section of the fetal abdomen at the level where the umbilical vein enters the liver. We used the mean of three measurements, and only one measurement was taken of each woman at 15–25 weeks' gestation. As gestational age is a determinant for AC and maternal weight, we used a z-score for AC and weight gain per week for the analysis. Z-score statistics and linear regression analysis were used to assess the effect of maternal weight gain in pregnancy on AC and birthweight. We used SPSS (Statistical Package for the Social Sciences; Inc, Chicago, IL, USA).

Table 1. Characteristics of the included participants (n=515).

Pre-pregnancy weight (kg)	65 (43–132)
Weight gain (kg)	14 (−5–34)
Height (cm)	168 (152–183)
BMI (body mass index)	23 (16–48)
Age (years)	30 (19–43)
Smokers (n)	41 (8%)

Values are median (range) or n (%).

Results

Of the 650 women included, 135 were excluded, leaving 515 for the statistical calculation. Four participants were excluded because of withdrawal from the study, a discrepancy between ultrasound and menstrual age of>14 days led to eight exclusions, and four women experienced miscarriage or intrauterine fetal death. Missing data on the AC measurements at gestational week 15–25 or missing data of weight at the last antenatal visit after gestational week 32 also led to exclusion of, respectively, 24 and 101 women. In six cases there were overlapping causes of exclusion.

Maternal characteristics of the participants are presented in Table 1. Gestational weeks at the last antenatal visit ranged from 32^{+0} to 41^{+6}. Table 2 presents the distribution of the last weight measurements according to gestational age. Mean weight gain was 14.5kg and mean weight gain per week was 0.39kg. There were 230 women who gained more than 0.40kg per week. In this group there was no significant association to fetal AC (p=0.829), but in the group of 96 women who gained less than 0.28kg per week, there were significantly lower AC measurements (p=0.000).

Linear regression with the z-score for AC as the dependent variable and the z-score for maternal weight gain (kg/week) as the independent variable (Figure 1), showed that maternal weight gain during pregnancy was positively associated to the fetal AC in the second trimester (r=0.122 (95%CI 0.051–0.194; p=0.001)). There was no significant association between pre-pregnant weight and AC or between BMI and AC between 15 and 25 weeks (Table 3).

Table 2. Distribution of the last weight measurement according to gestational age.

Gestational age (weeks+days)	n
32+0–33+6	55
34+0–35+6	99
36+0–37+6	165
38+0–39+6	166
40+0–	30

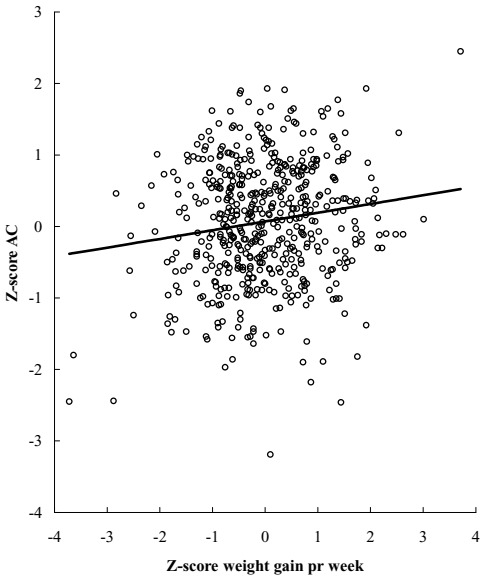


Figure 1. Z-score statistics showing a positive association between maternal weight gain per week in pregnancy and fetal abdominal circumference (AC), p= 0.001. Z-score AC=0.070+0.122 * (z-score of weight gain per week).

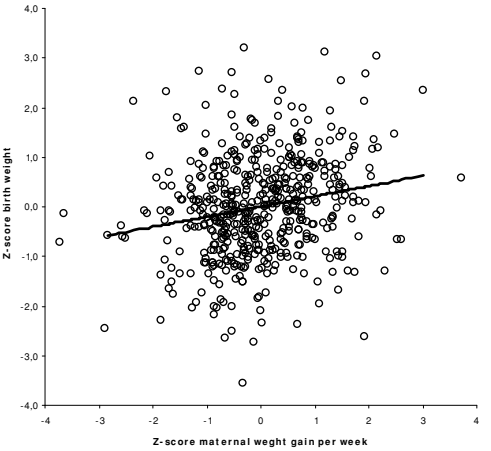


Figure 2. Z-score statistics showing a positive association between maternal weight gain pr week in pregnancy and birthweight, p=0.000. Z-score BW=−0.003+0.024 * (z-score of weight gain per week).

Mean birthweight was 3 723g (range 2 200g–5 450g). Maternal weight gain in pregnancy was positively associated with birthweight (p=0.000) (Figure 2).

Discussion

Our findings demonstrated a positive association between maternal weight gain in pregnancy and the fetal AC at 15–25 weeks' gestation, suggesting that maternal nutrition may impact on the fetal liver and influence growth from an early stage of fetal development. The effect seemed strongest in mothers with the slowest weight gain rate. This early association between weight gain and fetal size is in contrast to the findings of a previous study where only a low, although significant, association from gestational week 25 onwards was shown (4). The design of the study was different, as the effect of maternal risk factors in a population where such factors were prevailing was examined, whereas our study assessed growth dynamics in a carefully selected low-risk population. The populations were comparable concerning BMI but differed regarding ethnicity (69% black compared with our 0%), inclusion of smokers (49% compared with our 8%) and mothers having hypertension (6.4% compared with our 0%). These factors are known to influence fetal growth. Possibly more important would have been the differences in assignment of gestational age. Whereas the present study exclusively used the exact information of a regular LMP with cycle variation within 28±4 days to select a homogeneous population, Goldenberg et al. used LMP or ultrasound-corrected gestational age for an unknown proportion of their population, which may have affected their results concerning AC at 18 weeks' gestation. We regard our finding as an expression of nutritional mechanisms operating in physiological pregnancies. Several

Table 3. Effect of maternal pregnancy weight gain, pre-pregnancy weight and BMI on fetal abdominal circumference at 15–25 weeks' gestation. R, 95%CI and standard error (SE) expressed as a z-score.

	n	*r*	95%CI	SE	*p*-value
Weight gain	515	0.122	0.051–0.194	0.036	0.001
Weight gain>0.40kg/week	230	0.020	–0.160 to 0.199	0.091	0.829
Weight gain<0.28kg/week	96	0.554	0.261–0.846	0.147	0.000
Pre-pregnancy weight	515	÷0.002	–0.008 to 0.004	0.003	0.483
Pre-pregnancy BMI	515	÷0.012	–0.029 to 0.005	0.009	0.171

prior studies have demonstrated an association between maternal weight gain in pregnancy and fetal size from mid-pregnancy onwards, but not the exact effect of the different biometric parameters, and not as early as from gestational week 15 (3).

The fetal nutritional supply depends on maternal–placental transport capacity, maternal body composition and diet (18,19). Maternal weight gain may influence the fetus in several ways. The umbilical vein directs 75–80% of its nutrient-rich blood from the placenta to the fetal liver (20) but this fraction is reduced in cases of fetal growth restriction (21) leading to less hepatocyte proliferation (7). It has also been shown that low pregnancy weight gain is associated with reduced umbilical venous perfusion to the fetal liver, particularly the right lobe (22). A liver-sparing mechanism comes into play in slim mothers with low fat stores (6). Such adaptive measures are believed to alter hepatic development (23), with potential long-term consequences for the risk of cardiovascular and metabolic disease (6). We assume that such dynamics underlie the variation in liver size and thus AC.

The recently shown effect of umbilical flow and maternal nutritional status on fetal liver development (6,7,22) was demonstrated during the latter part of pregnancy. However, there is increasing evidence of lasting fetal growth differentiation before 24 weeks' gestation (17,24). Rather than using birthweight as a marker of later health risk, it may be of value to explore more specific fetal measurements. The AC offers such a possibility as a reflection of liver size and central fat accretion.

There was no association between maternal pre-pregnancy weight or BMI and fetal AC prior to gestational week 25. This is in agreement with a prior study (4).

A possible confounder in the present study was the registration of maternal weight. Pre-pregnancy weight was based on information from the antenatal chart. This was self-reported weight or measurement of weight at the first antenatal visit. The use of self-reported weight is associated with a tendency to underestimate the weight, by an average 1.4kg and more in heavy women (25). In a recent study of 1 000 women, it was shown that weight, BMI, water content and fat did not change during the first 14 weeks of pregnancy (26). We therefore assume that weight and body composition in early pregnancy within our study population was fairly similar to pre-pregnancy values. We chose weight at the last antenatal visit instead of weight at birth because few women were weighed on the day they gave birth. At the last antenatal visit we had a more accurate weight measurement.

There is increasing evidence that fetal size at second trimester is linked to neonatal and adult morbidity, and that maternal anthropometrical and nutritional status are determinants of fetal growth and adaptation (6,18,22). The present study adds the information that such regulation can be traced already by the second trimester, i.e. maternal weight gain in pregnancy impacts on fetal abdominal circumference, particularly when maternal weight gain is slow.

Funding

No specific funding.

Acknowledgements

Henriette Hellebust and Synnøve Lian Johnsen are supported by research grants from Haukeland University Hospital, Bergen, Norway. We thank midwife Rita Sollien for assistance with the ultrasound measurements.

References

1. Gluckman PD, Hanson MA. Maternal constraint of fetal growth and its consequences. Semin Fetal Neonatal Med. 2004;9:419–25.
2. Ounsted M, Scott A, Ounsted C. Transmission through the female line of a mechanism constraining human fetal growth. Int J Epidemiol. 2008;37:245–50.
3. Ay L, Kruithof CJ, Bakker R, Steegers EA, Witteman JC, Moll HA, et al. Maternal anthropometrics are associated with fetal size in different periods of pregnancy and at birth. The Generation R Study. Br J Obstet Gynaecol. 2009;116: 953–63.
4. Goldenberg RL, Davis RO, Cliver SP, Cutter GR, Hoffman HJ, Dubard MB, et al. Maternal risk factors and their influence on fetal anthropometric measurements. Am J Obstet Gynecol. 1993;168:1197–203; discussion 203–5.
5. Johnsen SL, Rasmussen S, Wilsgaard T, Sollien R, Kiserud T. Longitudinal reference ranges for estimated fetal weight. Acta Obstet Gynecol Scand. 2006;85:286–97.
6. Haugen G, Hanson M, Kiserud T, Crozier S, Inskip H, Godfrey KM. Fetal liver-sparing cardiovascular adaptations linked to mother's slimness and diet. Circ Res. 2005;96:12–14.
7. Tchirikov M, Kertschanska S, Sturenberg HJ, Schroder HJ. Liver blood perfusion as a possible instrument for fetal growth regulation. Placenta. 2002;23(Suppl A):S153–8.
8. Ebbing C, Rasmussen S, Godfrey KM, Hanson MA, Kiserud T. Redistribution pattern of fetal liver circulation in intrauterine growth restriction. Acta Obstet Gynecol Scand. 2009;88:1118–23.
9. Kessler JRS, Godfrey K, Hanson M, Kiserud T. Venous liver blood flow and regulation of human fetal growth: evidence from macrosomic fetuses. Am J Obstet Gynecol. 2011 (in press).
10. Boito SM, Struijk PC, Ursem NT, Stijnen T, Wladimiroff JW. Assessment of fetal liver volume and umbilical venous volume flow in pregnancies complicated by insulin-dependent diabetes mellitus. Br J Obstet Gynaecol. 2003;110:1007–13.

11. Committee on Nutritional Status During Pregnancy and Lactation IoM. Nutrition During Pregnancy: Part I: Weight Gain, Part II: Nutrient Supplements. Washington, DC: National Academy of Sciences, 1990. Available from: http://www.nap.edu/catalog/1451.html (accessed March 31, 2011).
12. Seidman DS, Ever-Hadani P, Gale R. The effect of maternal weight gain in pregnancy on birth weight. Obstet Gynecol. 1989;74:240–6.
13. Sekiya N, Anai T, Matsubara M, Miyazaki F. Maternal weight gain rate in the second trimester are associated with birth weight and length of gestation. Gynecol Obstet Invest. 2007;63:45–8.
14. Abrams B, Selvin S. Maternal weight gain pattern and birth weight. Obstet Gynecol. 1995;86:163–9.
15. Johnsen SL, Rasmussen S, Sollien R, Kiserud T. Fetal age assessment based on ultrasound head biometry and the effect of maternal and fetal factors. Acta Obstet Gynecol Scand. 2004;83:716–23.
16. Johnsen SL, Wilsgaard T, Rasmussen S, Sollien R, Kiserud T. Longitudinal reference charts for growth of the fetal head, abdomen and femur. Eur J Obstet Gynecol Reprod Biol. 2006;127:172–85.
17. Johnsen SL, Wilsgaard T, Rasmussen S, Hanson MA, Godfrey KM, Kiserud T. Fetal size in the second trimester is associated with the duration of pregnancy, small fetuses having longer pregnancies. BMC Pregnancy Childbirth. 2008;8:25.
18. Godfrey KM, Barker DJ. Fetal nutrition and adult disease. Am J Clin Nutr. 2000;71(5 Suppl):1344S–52S.
19. Lewis RM, Greenwood SL, Cleal JK, Crozier SR, Verrall L, Inskip HM, et al. Maternal muscle mass may influence system A activity in human placenta. Placenta. 2010;31:418–22.
20. Kiserud T, Rasmussen S, Skulstad S. Blood flow and the degree of shunting through the ductus venosus in the human fetus. Am J Obstet Gynecol. 2000;182(1 Pt 1):147–53.
21. Kiserud T, Kessler J, Ebbing C, Rasmussen S. Ductus venosus shunting in growth-restricted fetuses and the effect of umbilical circulatory compromise. Ultrasound Obstet Gynecol. 2006;28:143–9.
22. Kessler J, Rasmussen S, Godfrey K, Hanson M, Kiserud T. Longitudinal study of umbilical and portal venous blood flow to the fetal liver: low pregnancy weight gain is associated with preferential supply to the fetal left liver lobe. Pediatr Res. 2008;63:315–20.
23. Darp RA, de Boo HA, Phua HH, Oliver MH, Derraik JG, Harding JE, et al. Differential regulation of igf1 and igf1r mRNA levels in the two hepatic lobes following intrauterine growth restriction and its treatment with intra-amniotic insulin-like growth factor-1 in ovine fetuses. Reprod Fertil Dev. 2010;22:1188–97.
24. Rasmussen S, Kiserud T, Albrechtsen S. Foetal size and body proportion at 17–19 weeks of gestation and neonatal size, proportion, and outcome. Early Hum Dev. 2006;82:683–90.
25. Spencer EA, Appleby PN, Davey GK, Key TJ. Validity of self-reported height and weight in 4808 EPIC-Oxford participants. Public Health Nutr. 2002;5:561–5.
26. Fattah C, Farah N, Barry SC, O'Connor N, Stuart B, Turner MJ. Maternal weight and body composition in the first trimester of pregnancy. Acta Obstet Gynecol Scand. 2010;89:952–5.

Errata for
Maternal influence on fetal size and use of longitudinal fetal surveillance in predicting adverse perinatal outcomes

Henriette Odland Karlsen

Thesis for the degree philosophiae doctor (PhD)

at the University of Bergen

Henriette O. Karlsen

(signature of candidate)

(signature of faculty)

21.04.16

Errata

Page 22 "that maternal constrain" – Rettes til: "that maternal constraint"

Page 24 "education and birthweight is seen (97).Education is" Rettes til: "education and birthweight is seen (97). Education is"

Page 28 "***Figure 4.*** *Demonstration of central fetal circulation (reproduced by permission (150).*" Rettes til: "***Figure 4.*** *Demonstration of central fetal circulation (reproduced by permission (150)).*"

Page 32 "affected followed by MCA PI <5" Rettes til: "affected followed by MCA PI <5th centile"

Page 41 "on AC and birth weight" Rettes til: "on AC and birthweight"

Page 57 "were not registered in the MFR" Rettes til: "were not registered in the MBR"