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Performance of third-trimester combined screening model for prediction of adverse perinatal outcome

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KEYWORDS: biochemical markers; Doppler; fetal growth restriction; perinatal outcome; small-for-gestational age; third-trimester screening

ABSTRACT

Objective *To explore the potential value of third-trimester combined screening for the prediction of adverse perinatal outcome (APO) in the general population and among small-for-gestational-age (SGA) fetuses.*

Methods *This was a nested case–control study within a prospective cohort of 1590 singleton gestations undergoing third-trimester evaluation (32 + 0 to 36 + 6 weeks' gestation). Maternal baseline characteristics, mean arterial blood pressure, fetoplacental ultrasound and circulating biochemical markers (placental growth factor (PlGF), lipocalin-2, unconjugated estriol and inhibin A) were assessed in all women who subsequently had an APO (*n = *148) and in a control group without perinatal complications (*n = *902). APO was defined as the occurrence of stillbirth, umbilical artery cord blood pH < 7.15, 5-min Apgar score < 7 or emergency operative delivery for fetal distress. Logistic regression models were developed for the prediction of APO in the general population and among SGA cases (defined as customized birth weight < 10th centile).*

Results *The prevalence of APO was 9.3% in the general population and 27.4% among SGA cases. In the general population, a combined screening model including* a-priori *risk (maternal characteristics), estimated fetal weight (EFW) centile, umbilical artery pulsatility index (UA-PI), estriol and PlGF achieved a detection rate for APO of 26% (area under receiver–operating characteristics curve (AUC), 0.59 (95% CI, 0.54–0.65)), at a 10% false-positive rate (FPR). Among SGA cases, a model including* a-priori *risk, EFW centile, UA-PI, cerebroplacental ratio, estriol and PlGF predicted 62% of APO (AUC, 0.86 (95% CI, 0.80–0.92)) at a FPR of 10%.*

Conclusions *The use of fetal ultrasound and maternal biochemical markers at 32–36 weeks provides a poor prediction of APO in the general population. Although it remains limited, the performance of the screening model is improved when applied to fetuses with suboptimal fetal growth.* Copyright © 2016 ISUOG. Published by John Wiley & Sons Ltd.

INTRODUCTION

Despite a substantial improvement in obstetric care over the past decades, each year adverse perinatal outcomes (APOs) late in gestation contribute to almost one-quarter of the 3 million fetal and neonatal deaths documented worldwide[1–3]. While some APOs have their origins in access to and quality of healthcare[4], many more may be rooted in maternal health conditions and a complex interaction between placental dysfunction, hormonal regulation and fetal oxygenation, making their prediction difficult and limiting preventive action.

Surveillance of fetal wellbeing late in gestation has relied on different approaches, such as quantification of amniotic fluid, monitoring fetal movements and fetal cardiotocography[5,6]. Although the majority are relatively inexpensive, their widespread use has not resulted in a significant reduction in perinatal mortality and unfortunately, in some cases, has increased unnecessary interventions[7,8]. Another proposed strategy is the identification of pregnancies at a higher risk of APO, such as those with fetal growth restriction, through the use of fetoplacental ultrasound[9,10]. Assessment of fetal growth and umbilical artery (UA) Doppler have been shown to reduce perinatal mortality in high-risk pregnancies[11], an effect that has not been seen in low-risk populations[12].

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More recently, the cerebroplacental ratio (CPR) has been proposed by several authors as a useful marker for the prediction of APO near term and intrapartum fetal compromise[13–18]. Finally, placentally derived proteins associated with fetal death, growth restriction and fetal metabolic diseases, such as unconjugated estriol[19–23], placental growth factor (PlGF)[24–27], inhibin A[28–31] or lipocalin-2[32], have also been proposed as useful biomarkers for the prediction of APO associated with placental dysfunction[33].

Recent studies have shown that the prediction of APO at term or during labor by combining fetoplacental Doppler and circulating biomarkers is of limited utility[34,35]. However, the predictive ability of these models has not been differentiated between uncomplicated and high-risk pregnancies. The objective of this study was to explore the performance of a third-trimester combined model, including maternal characteristics, fetoplacental ultrasound and biochemical markers, for predicting APO in the general population and in small-for-gestational-age (SGA) fetuses in a nested case–control study within a cohort of 1590 women, which included 148 cases with an APO.

METHODS

Study population

This was a nested case–control study drawn from a large prospective cohort of 1590 women with a singleton pregnancy attending their routine hospital visit in the third trimester of pregnancy (32 + 0 to 36 + 6 weeks' gestation) at the Department of Maternal-Fetal Medicine in Hospital Clinic Barcelona between January 2012 and December 2014. This visit included recording baseline maternal characteristics, measurement of blood pressure, fetoplacental ultrasound and collection of maternal serum. The analysis of biomarkers was conducted in 1050 patients, which included all women who subsequently had an APO ($n = 148$) and a group of controls in a ratio of approximately 6:1 ($n = 902$) comprising consecutive uncomplicated pregnancies in the same period, matched for gestational age (GA) at scan (± 2 weeks). All control pregnancies delivered appropriate-for-gestational-age (AGA) neonates with a birth weight ≥ 10th centile according to local standards[36]. GA in all pregnancies was calculated on the basis of the measurement of fetal crown–rump length at 11–13 weeks. The institutional ethics committee approved the study protocol (IRB 2012/7154), and all patients provided written informed consent. The study protocol consisted of evaluation of maternal baseline characteristics, blood pressure, fetoplacental ultrasound and maternal biochemical markers at 32 + 0 to 36 + 6 weeks' gestation and subsequent recording of perinatal outcomes.

Predictive variables

Baseline characteristics

Maternal baseline characteristics, including demographic details and obstetric and medical histories, were recorded at the time of the third-trimester visit using a patient questionnaire, and data were entered into our database. The following variables were registered: maternal age, ethnicity, nulliparity (no previous delivery after 24 weeks of pregnancy), maternal height and weight, smoking during pregnancy (yes or no), method of conception (spontaneous or use of assisted reproductive technology), medical history (including chronic hypertension, diabetes mellitus, renal disease, autoimmune disease such as systemic lupus erythematous or antiphospholipid syndrome, congenital and acquired thrombophilic conditions) and obstetric history (including previous pregnancy complicated by pre-eclampsia, SGA or stillbirth). Maternal height and weight were measured at enrollment and body mass index calculated. Socioeconomic status was categorized into three levels: low (no education/educated to primary school level and/or no work), mid (educated to secondary school level and working) and high (attended university and working).

Third-trimester maternal blood pressure

Maternal blood pressure was measured automatically at 32 + 0 to 36 + 6 weeks' gestation with a calibrated OMRON M6 Confort device (OMRON Corporation, Kyoto, Japan), according to standard procedure. Blood pressure was measured in one arm (right or left) without distinction, while women were seated and after a 5-min rest. Mean arterial pressure (MAP) was calculated as: diastolic blood pressure + (systolic blood pressure – diastolic blood pressure)/3.

Third-trimester fetoplacental ultrasound

Transabdominal ultrasound with Doppler evaluation was performed with a 6–4-MHz probe (Siemens Sonoline Antares, Siemens Medical Systems, Malvern, PA, USA) and a Voluson 730 Expert Machine (GE Medical Systems, Zipf, Austria) including fetal biometry and fetoplacental Doppler. Estimated fetal weight (EFW) was calculated by means of the Hadlock formula using ultrasound measurement of the fetal head circumference, abdominal circumference and femur length[37]. EFW centile was calculated using local standards[36]. Doppler recordings were performed in the absence of fetal movement and voluntarily suspended maternal breathing. The examination at enrollment included: UA pulsatility index (PI) calculated from three or more consecutive waveforms obtained from a free-floating portion of the umbilical cord, at insonation angles of $<30°$[38]. Fetal middle cerebral artery (MCA) flow velocity waveforms were recorded at 1–2 cm from the circle of Willis, at insonation angles of less than 30°. CPR was calculated as the ratio of MCA-PI to UA-PI[13]. For uterine artery (UtA) evaluation, the probe was placed on the lower quadrant of the abdomen, angled medially, and color Doppler imaging was used to identify the UtA at the apparent crossover with the external iliac artery. Mean UtA-PI was calculated as the average PI of the right and left arteries[39].

Third-trimester maternal blood biomarkers

Maternal venous blood samples were collected in serum tubes and processed within 1 h. Serum was separated by centrifugation at 3000 rpm for 10 min at 4°C, and samples were stored immediately at −80°C until assayed. Serum concentrations of PlGF, unconjugated estriol, inhibin A and lipocalin-2 were measured using the AutoDELFIA® automated immunoanalyzer (PerkinElmer, Turku, Finland). Samples were assigned randomly to each plate, and assays for cases and controls were always run in parallel. Biochemical markers were measured with time-resolved fluorescence immunoassays (DELFIA) on an automated platform. For PlGF, unconjugated estriol and inhibin A, we used commercial AutoDELFIA® kits (PerkinElmer) and, for lipocalin-2, research reagents were prepared for this study. The AutoDELFIA PlGF kit had a measuring range from 5.6 pg/mL to 4000 pg/mL and a run control coefficient of variation (CV%) of 3.2%. The AutoDELFIA unconjugated estriol kit had a measuring range from 0.2 nmol/L to 50 nmol/L and a run control CV% of < 3.7%. The AutoDELFIA inhibin A kit had a measuring range from 8.0 pg/mL to 2000 pg/mL and a run control CV% of 3.9%. Lipocalin-2 assay was performed using monoclonal capture antibody (MAB17571, R&D Systems, Abingdon, UK) and polyclonal tracer antibody (AF1757, R&D Systems). Lipocalin-2 research assay had a measuring range from 0.2 ng/mL to 6.0 ng/mL and a run control CV% of 2.0%. Samples for the lipocalin-2 assay were diluted 1:100. Calibrators and quality (run) controls for all assays were run in duplicates on each plate and serum samples in singles. The laboratory personnel were blinded to the clinical results or the outcomes of the patients.

Perinatal outcome

Perinatal outcome was ascertained at delivery by reviewing medical records. APO in the general population and among SGA newborns was considered the main outcome of this study and was defined as the occurrence of stillbirth, emergency operative delivery (vaginal operative delivery or Cesarean section) owing to non-reassuring fetal status, low Apgar score or the presence of neonatal metabolic acidosis. Non-reassuring fetal status was defined as an abnormal fetal heart rate tracing and abnormal fetal scalp blood pH during intrapartum monitoring[5]. Briefly, continuous fetal heart rate monitoring was performed and tracings were classified as normal, suspicious or abnormal, according to the presence, type and length of decelerations, bradycardia, tachycardia and assessment of variability[5]. In cases with two or more criteria of suspicion and one or more criteria of abnormality not responding to fetal scalp digital stimulation, fetal scalp blood sampling was attempted and considered abnormal if pH was < 7.20. Apgar score was considered to be low if it was < 7 at 5 min. Neonatal metabolic acidosis was defined as a UA pH of < 7.15 and a base excess of > 12 mEq/L in the newborn[40]. All cases with adverse outcome were evaluated in a confidential enquiry to assure adherence to such guidelines. APO was considered in both the general population and among SGA newborns.

Statistical analysis

Student's *t*-test or the Mann–Whitney *U*-test and Pearson's chi-square test were used to perform univariate comparisons between groups of quantitative and qualitative variables, respectively. Categorical data are presented as *n* (%) and continuous data as median (interquartile range (IQR)). EFW and Doppler measurements were expressed as the respective percentile and *Z*-score, adjusted for GA. Values of MAP, PlGF, unconjugated estriol, inhibin A and lipocalin-2 were $\log_{10}$ transformed to make their distribution Gaussian, then each value was expressed as a multiple of the normal median (MoM) after adjustment for characteristics that provided a substantial contribution to the log-transformed value.

In each patient, the *a-priori* risk for APO was calculated using multivariable logistic regression analysis with backward stepwise elimination by sequentially removing non-significant ($P > 0.05$) variables to determine which of the factors among maternal characteristics had a significant contribution to predicting APO. The performance for the prediction of APO by *a-priori* risk ($\log_{10}$), MAP, EFW centile, UtA-PI, UA-PI, CPR, PlGF, unconjugated estriol, inhibin A and lipocalin-2, individually and in various combinations, was determined by receiver–operating characteristics (ROC) curve analysis. The resulting areas under the ROC curves (AUCs) were compared using the DeLong method, and $P < 0.05$ was considered to be statistically significant. Finally, detection rates were calculated for a 10% false-positive rate (FPR). Statistical analysis was performed using SPSS Statistics version 20 (SPSS Inc., Chicago, IL, USA), and STATA 14 (StataCorp LP, 2015, College Station, TX, USA).

RESULTS

Prediction of adverse perinatal outcome in overall population

Among the 1590 patients who were evaluated, 148 (9.3%) gave birth to neonates with an APO (four stillbirths, 69 cases of fetal distress requiring emergency operative delivery, 76 cases of neonatal acidosis and 29 with a low Apgar score, some cases having more than one adverse event) and were matched with 902 controls who delivered newborns with no perinatal complications. Clinical characteristics, maternal and perinatal outcomes and sonographic and biochemical results were obtained for all patients and are shown in Table 1. There were no significant differences with respect to maternal age, body mass index, ethnicity or incidence of autoimmune disease. In the APO group, median values of EFW centile were significantly lower than in the control group. Furthermore, the median *Z*-score value of mean UA-PI was significantly higher in APO cases, while there was no difference in UtA-PI and

Table 1 Maternal baseline, pregnancy and perinatal characteristics of uncomplicated pregnancies, pregnancies with adverse perinatal outcome (APO) and pregnancies with small-for-gestational-age (SGA) fetuses and APO

Characteristic	*Uncomplicated pregnancies* (n = *902*)	*Pregnancies with APO* (n = *148*)	P*	*SGA pregnancies with APO* (n = *48*)	P†
Maternal baseline characteristic					
Age (years)	32 (28 to 36)	32 (28 to 35)	0.78	34 (30 to 38)	0.05
BMI (kg/m^2)	22 (20 to 25)	22 (20 to 25)	0.45	22 (20 to 25)	0.67
Ethnicity			0.77		0.03
White	591 (65.5)	93 (62.8)		37 (77.1)	
Latin	201 (22.3)	35 (23.6)		5 (10.4)	
Other	110 (12.2)	20 (13.5)		6 (12.5)	
Nulliparous	561 (62.2)	84 (56.8)	0.21	27 (56.3)	0.41
Smoker during pregnancy	84 (9.3)	17 (11.5)	0.65	12 (25.0)	0.002
Chronic hypertension	7 (0.8)	3 (2.0)	0.15	2 (4.2)	0.02
Diabetes	36 (4.0)	4 (2.7)	0.5	0 (0)	0.73
Autoimmune disease	9 (1.0)	4 (2.7)	0.07	2 (4.2)	0.13
Assisted reproductive technology	27 (3.0)	1 (0.7)	0.1	0 (0)	0.22
Previous history of SGA	13 (1.4)	2 (1.4)	0.9	1 (2.1)	0.89
Parameters at third-trimester evaluation					
GA at evaluation (weeks)	33.6 (33.0 to 34.1)	33.4 (33.0 to 33.8)	0.06	34 (33 to 34.5)	0.36
Mean maternal BP (mmHg)	83 (76 to 88)	83 (76 to 91)	0.14	93 (78 to 114)	0.001
Estimated fetal weight (g)	2177 (2029 to 2353)	2109 (1944 to 2250)	< 0.001	1913 (1571 to 2152)	< 0.001
Estimated fetal weight centile	50 (32 to 71)	47 (18 to 69)	0.03	10 (1 to 35)	< 0.001
Mean uterine artery PI *Z*-score	−0.48 (−1.16 to 0.28)	−0.44 (−1.12 to 0.63)	0.24	0.30 (−0.67 to 2.39)	< 0.001
Umbilical artery PI *Z*-score	−0.22 (−0.53 to 0.16)	0.39 (−0.50 to 0.49)	0.003	0.57 (−0.60 to 1.56)	< 0.001
Middle cerebral artery PI *Z*-score	0.1 (−0.33 to 1)	0.1 (−0.30 to 0.9)	0.90	−0.2 (−0.4 to 0.8)	0.61
Cerebroplacental ratio *Z*-score	−0.25 (−1.0 to 0.56)	−0.51 (−1.3 to 0.61)	0.05	−1.42 (−2.22 to −0.43)	< 0.001
Maternal serum estriol MoM	1.00 (0.95 to 1.06)	0.98 (0.9 to 1.06)	0.03	0.9 (0.65 to 1.01)	< 0.001
Maternal serum PlGF MoM	1.00 (0.89 to 1.10)	0.93 (0.82 to 1.09)	0.001	0.79 (0.58 to 0.96)	< 0.001
Maternal serum lipocalin-2 MoM	1.00 (0.95 to 1.06)	1.01 (0.90 to 1.07)	0.12	1.06 (0.98 to 1.10)	< 0.001
Maternal serum inhibin A MoM	1.00 (0.95 to 1.04)	1.00 (0.95 to 1.06)	0.29	1.03 (0.97 to 1.11)	0.005
Pregnancy and perinatal outcome					
GA at delivery (weeks)	39.9 (38.9 to 40.7)	40.0 (38.5 to 40.6)	0.7	38.7 (34.3 to 40.3)	< 0.001
Induction of labor	233 (25.8)	50 (33.8)	0.04	22 (45.8)	0.002
Cesarean section	169 (18.7)	80 (54.1)	< 0.001	32 (66.7)	< 0.001
Birth weight (g)	3300 (2997 to 3580)	3100 (2748 to 3500)	< 0.001	2505 (1612 to 2796)	< 0.001
Birth-weight percentile	43 (19 to 72)	24 (4 to 60)	< 0.001	2 (0 to 4)	< 0.001
Pre-eclampsia	28 (3.1)	14 (9.5)	< 0.001	13 (27.1)	< 0.001
Cesarean delivery for NRFS	0 (0)	65 (43.9)	< 0.001	25 (52.1)	< 0.001
5-min Apgar score < 7	0 (0)	29 (19.6)	< 0.001	19 (39.6)	< 0.001
Neonatal acidosis	0 (0)	76 (51.4)	< 0.001	17 (35.4)	< 0.001
Fetal death	0 (0)	4 (2.7)	< 0.001	1 (2.1)	0.05

Data are presented as *n* (%) or median (interquartile range). Comparison of uncomplicated pregnancies with: *pregnancies with APO; †SGA pregnancies with APO. BMI, body mass index; BP, blood pressure; GA, gestational age; MoM, multiples of the median of log-transformed value; NRFS, non-reassuring fetal status; PI, pulsatility index; PlGF, placental growth factor.

CPR Z-scores compared with the control group. Moreover, compared to the control group, the mean $\log_{10}$ maternal serum MoM concentrations of PlGF and estriol were significantly lower in the APO group, while there was no significant difference in the median $\log_{10}$ maternal serum MoM concentrations of lipocalin-2 or inhibin A. Although there was no significant difference in GA at delivery between cases and controls, birth-weight centile was significantly lower in cases with APO than in controls. The rates of pre-eclampsia and obstetric intervention, such as induction of labor and Cesarean section, were significantly higher in the APO group than in controls (Table 1).

Adjusted odds ratios (ORs) of each maternal factor in the prediction algorithms for APO are presented in Table S1. The likelihood of APO was not affected significantly by maternal age ($P = 0.59$), ethnicity ($P = 0.45$), body mass index ($P = 0.35$) or smoking ($P = 0.4$). Multivariable regression analysis demonstrated that, in the prediction of APO, there were significant independent contributions from EFW centile (OR, 0.99 (95% CI, 0.98–0.99), $P = 0.02$), UA-PI (OR, 1.58 (95% CI, 1.28–1.94), $P < 0.001$) and CPR (OR, 0.82 (95% CI, 0.69–0.97), $P = 0.02$), as well as maternal serum concentrations of estriol (OR, 0.97 (95% CI, 0.96–0.98), $P < 0.001$) and PlGF (OR, 0.98 (95% CI, 0.97–0.99), $P < 0.001$), but not inhibin A (OR, 1.02 (95% CI, 0.99–1.04), $P = 0.1$) or lipocalin-2 (OR, 1.02 (95% CI, 0.99–1.04), $P = 0.06$). The best prediction for APO was provided by a combination of *a-priori* risk (including chronic hypertension and socioeconomic status), EFW centile, UA-PI, unconjugated estriol and PlGF, achieving a detection rate of 26% at a FPR of 10%. AUCs of the *a-priori* risk, EFW centile, Doppler indices and biochemical markers, and their

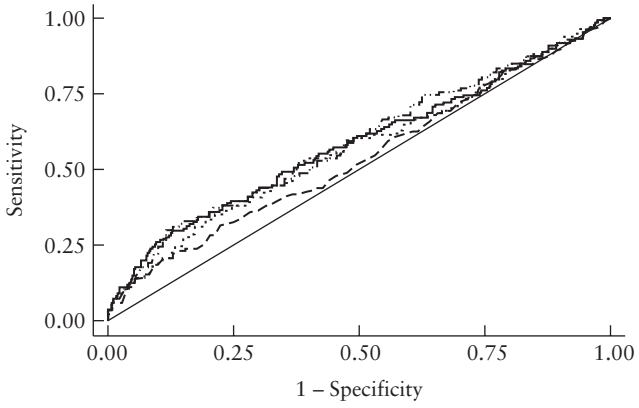

Figure 1 Receiver–operating characteristics curves for prediction of adverse perinatal outcome in general population by estimated fetal weight (EFW) centile (– – – –), combination of estriol and placental growth factor (PlGF) (–··–··-), combination of EFW centile, estriol and PlGF (·····) and combination of maternal baseline characteristics, EFW centile, umbilical artery pulsatility index, estriol and PlGF (———).

Table 2 Screening performance for detection of adverse perinatal outcome in general population

Screening test	*AUC (95% CI)*	*DR (%) at 10% FPR*
Maternal *a-priori* risk*	0.547 (0.50–0.59)	2
EFW centile	0.537 (0.48–0.59)	19
EFW centile + estriol + PlGF	0.568 (0.51–0.62)	20
Maternal *a-priori* risk*plus:		
EFW centile	0.565 (0.51–0.62)	19
UA-PI	0.552 (0.50–0.61)	18
CPR	0.555 (0.50–0.61)	17
Estriol	0.563 (0.50–0.61)	19
PlGF	0.583 (0.53–0.64)	21
EFW centile + UA-PI	0.565 (0.51–0.62)	20
Estriol + PlGF	0.587 (0.53–0.64)	24
EFW centile + UA-PI + estriol + PlGF	0.589 (0.54–0.65)	26

*Including chronic hypertension and socioeconomic status.
AUC, area under receiver–operating characteristics curve; CPR, cerebroplacental ratio; DR, detection rate; EFW, estimated fetal weight; FPR, false-positive rate; PI, pulsatility index; PlGF, placental growth factor; UA, umbilical artery.

combination, for the prediction of APO are illustrated in Figure 1, with AUC results summarized in Table 2.

Prediction of adverse perinatal outcome among SGA fetuses

Among the study population, 175 fetuses had a birth weight < 10th centile and were categorized as SGA. Among these, 48 (27.4%) cases had an APO (one fetal death, 25 cases of fetal distress requiring emergency Cesarean section, 17 cases of neonatal acidosis and 19 neonates with a low Apgar score, some cases having more than one adverse event). The epidemiological and clinical characteristics of SGA cases with APO are shown in Table 1. The prevalence of smoking during pregnancy was significantly higher in the group with a SGA fetus with APO than in the control group. Similarly, the prevalence of chronic hypertension was significantly higher among the SGA pregnancies with APO. Among SGA pregnancies with APO, median *Z*-score values of mean UtA-PI and UA-PI were significantly higher, while the *Z*-score of CPR was significantly lower than in controls. Moreover, compared with the control group, the mean $\log_{10}$ maternal serum MoM concentrations of PlGF and estriol were significantly lower and of lipocalin-2 and inhibin A were significantly higher in the group of SGA cases with APO. The rate of obstetric intervention (induction of labor and Cesarean section) was significantly higher in the group of SGA cases with APO, as was the incidence of pre-eclampsia (Table 1).

Multivariable regression analysis demonstrated that, for the prediction of APO in SGA fetuses, there were significant independent contributions to the *a-priori* risk from maternal characteristics such as age (OR, 1.05 (95% CI, 1.01–1.11), $P = 0.04$), smoking (OR, 3.41 (95% CI, 1.71–6.80), $P < 0.001$) and chronic hypertension (OR, 5.40 (95% CI, 1.11–26.10), $P = 0.04$). Furthermore, EFW centile (OR, 0.95 (95% CI, 0.93–0.96), $P < 0.001$), UA-PI (OR, 2.85 (95% CI, 2.09–3.87), $P < 0.001$), CPR (OR, 0.37 (95% CI, 0.27–0.50), $P < 0.001$) and mean UtA-PI (OR, 1.81 (95% CI, 1.49–2.19), $P < 0.001$) were significant independent contributors to the prediction of APO in SGA cases (Table S1), as well as maternal serum biomarkers estriol (OR, 0.95 (95% CI, 0.93–0.96), $P < 0.001$), PlGF (OR, 0.94 (95% CI, 0.92–0.96), $P < 0.001$), lipocalin-2 (OR, 1.07 (95% CI, 1.03–1.11), $P < 0.001$) and inhibin A (OR, 1.06 (95% CI, 1.03–1.10), $P = 0.001$).

AUCs of the *a-priori* risk, EFW centile, Doppler indices and biochemical markers, and their combination, for the prediction of APO in SGA cases are shown in Figure 2, with AUC results summarized in Table 3. The best prediction model for APO in SGA cases was provided by a combination of *a-priori* risk (including chronic hypertension and low socioeconomic status), EFW centile, UA-PI, CPR, PlGF and estriol, achieving a detection rate of 62% at a 10% FPR (AUC, 0.86 (95% CI, 0.80–0.92)) (Table 3 and Figure 2). Importantly, the performance of the model was similar to that obtained by a combination of maternal characteristics, EFW centile and UA-PI or CPR, without biochemical markers, predicting, at a 10% FPR, 64% (AUC, 0.83 (95% CI, 0.76–0.90)) and 63% (AUC, 0.84 (95% CI, 0.77–0.90)) of APO in SGA cases, respectively.

DISCUSSION

This study provides evidence that a combined screening model based on maternal characteristics, fetoplacental Doppler and biochemical markers has poor performance for the prediction of APO in the general population, while its predictive performance is better when applied to pregnancies with suboptimal fetal growth.

Our results are comparable with those reported by two recent studies from The Fetal Medicine Foundation describing the prediction of APO in the third trimester[34,35]. The authors reported that, at 30–34 weeks'

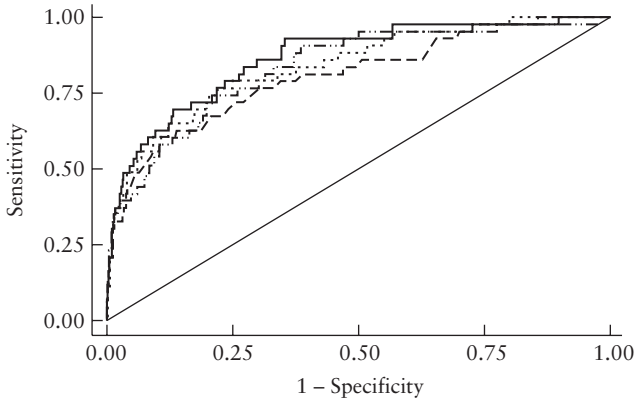

Figure 2 Receiver–operating characteristics curves for prediction of adverse perinatal outcome in small-for-gestational-age fetuses by estimated fetal weight (EFW) centile (– – – –), combination of estriol and placental growth factor (PlGF) (–··–··-), combination of EFW centile, estriol and PlGF (·····) and combination of maternal baseline characteristics, umbilical artery pulsatility index, cerebro-placental ratio, estriol and PlGF (———).

Table 3 Screening performance for detection of adverse perinatal outcome in small-for-gestational-age fetuses

Screening test	*AUC (95% CI)*	*DR (%) at 10% FPR*
Maternal *a-priori* risk*	0.701 (0.61–0.79)	39
EFW centile	0.804 (0.73–0.88)	56
EFW centile + estriol + PlGF	0.839 (0.77–0.90)	60
Maternal *a-priori* risk*plus:		
MAP	0.754 (0.66–0.85)	42
EFW centile	0.817 (0.75–0.89)	56
UA-PI	0.785 (0.71–0.86)	52
CPR	0.797 (0.72–0.87)	53
Estriol	0.793 (0.72–0.87)	46
PlGF	0.797 (0.72–0.87)	46
Estriol + PlGF	0.839 (0.77–0.90)	53
EFW centile + CPR	0.837 (0.77–0.90)	63
EFW centile + UA-PI + CPR + estriol + PlGF	0.862 (0.80–0.92)	62

*Including chronic hypertension and socioeconomic status.
AUC, area under receiver–operating characteristics curve; CPR, cerebroplacental ratio; DR, detection rate; EFW, estimated fetal weight; FPR, false-positive rate; MAP, mean arterial pressure; PI, pulsatility index; PlGF, placental growth factor; UA, umbilical artery.

gestation, the combination of MAP, fetal biometry, feto-placental Doppler (UtA, UA and MCA) and angiogenic factors (PlGF and soluble fms-like tyrosine kinase-1) provides good predictive performance for preterm pre-eclampsia and preterm SGA, as well as for fetal distress before labor. However, the performance of the model was poor for the prediction of stillbirth and adverse events during labor[35]. Similarly, a combined model at 35–37 weeks was found to be good for the prediction of pre-eclampsia, SGA and fetal distress before labor, but not for adverse events during labor[34]. Our study focused on APO, defined as stillbirth or adverse events during labor, and, although a new set of biochemical markers (lipocalin-2, inhibin A, estriol) was combined with Doppler parameters, the performance of the model was not improved. A potential explanation for this is that 32–36 weeks is too early to identify placental insufficiency or chronic fetal hypoxia susceptible to developing acute adverse events during labor.

When biomarkers were analyzed separately, EFW centile and UA Doppler were acceptable methods for the prediction of APO in the general population. However, this improvement does not seem to be clinically relevant. A possible explanation for the limited performance of fetal biometry is that the majority of APO occurs among non-SGA infants[41,42]. Similarly, the limited predictive performance observed for CPR is in agreement with recent studies reporting that the prediction of APO using the CPR at 32, 36 or 37 weeks is poor[42–44]. Nevertheless, it is possible that the predictive ability of CPR may be determined by the time lapse from evaluation to event, as other studies have reported that, close to delivery, CPR is independently associated with operative delivery, admission to the neonatal intensive care unit, low neonatal pH and emergency Cesarean section[16–18]. Likewise, inclusion of UtA Doppler did not improve the predictive value of the models. It is probable that increased UtA impedance, which indirectly reflects abnormal trophoblastic invasion, is not a major contributor to the majority of adverse intrapartum events, explaining its limited role in the combined models.

The biochemical markers employed in this study have been proposed as early potential markers of pregnancy complications. Although many of the associations were statistically significant in the univariate analysis as well as in previous studies[23,27,32,45–49], the sensitivity and positive predictive values for the individual outcomes were relatively low. The combination of estriol and PlGF was only a modest predictor of these outcomes in our population. It is probable that both fetoplacental Doppler and the biochemical markers employed in the model reflect the same process (placental dysfunction), which may explain why there is not an additive effect in the performance of the model when they are combined.

When the combined model was applied to SGA fetuses, the predictive performance for APO was significantly improved, to a detection rate of 62%. In this subgroup, EFW centile, UA Doppler and CPR were able to recognize fetuses at a higher risk of APO. These findings are in line with those of previous studies demonstrating that brain sparing in SGA fetuses is associated with poorer perinatal outcome and higher risk of Cesarean delivery for non-reassuring fetal status[50,55]. Indeed, our group have reported recently that identification of small fetuses at risk of APO is possible using a combination of EFW centile, UtA Doppler and CPR[56,57].

In terms of biochemical markers, again, the combination of estriol and PlGF was of significant utility in the predictive performance of the model in SGA fetuses. We have also reported previously that, in SGA fetuses, angiogenic factors at diagnosis and follow-up with Doppler ultrasound have an acceptable performance (AUC, 0.68) for the prediction of APO[27]. Other authors have reported

that low maternal serum concentrations of estriol in the second trimester are associated with an increased risk of APO in SGA fetuses[45]. It is also important to note that, in both the general population and SGA fetuses, the predictive performance of biochemical markers was similar to that obtained by fetoplacental Doppler. This result is relevant, because in low-resource countries, there is a limitation on the widespread use of ultrasound because of the demands of the equipment and human resources, and biochemical markers may help to identify patients at risk who require referral to a tertiary center.

Strengths and limitations of the study

Strengths of this study are the inclusion of a well-defined cohort including both AGA and SGA pregnancies, defined according to fetal customized centiles, with very few losses to follow-up, meaning that the possibility of selection bias was minimized. Obstetricians in charge of labor monitoring were blinded to the results of the final combined model, which means that decisions in the labor and delivery room were not modified by the results of the combined screening. We acknowledge that, although the design of this study is an efficient way to explore potential predictors, nested case–control studies are susceptible to bias, and the performance of the model should be validated in other populations. The biggest weakness of this study is that labor is a big confounder. During labor, the fetoplacental relationship is tested to its highest degree and even if a fetus is not compromised before entering labor, intrapartum events can influence the possibility of an APO, which will not be predicted antepartum. Another limitation is the use of composite outcomes. It is possible that the outcomes may overlap and their merging may distort the predictive performance and not allow us to discern the performance of the model for each of the specific outcomes.

Clinical relevance

A combination of maternal characteristics, fetoplacental ultrasound and maternal biochemical markers at 32–36 weeks' gestation provides poor prediction of APO. The prediction of APO is challenging owing to its multiple causality and late occurrence in pregnancy and, therefore, future studies addressing new biomarkers and/or delaying the time of screening are warranted. In order to build effective screening models for the prediction of APO, understanding the polymorphic nature of antepartum and intrapartum adverse outcomes and the underlying pathophysiology is required.

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REFERENCES

1. Lawn JE, Blencowe H, Waiswa P, Amouzou A, Mathers C, Hogan D, Flenady V, Frøen JF, Qureshi ZU, Calderwood C, Shiekh S, Jassir FB, You D, McClure EM, Mathai M, Cousens S; Lancet Ending Preventable Stillbirths Series study group; Lancet Stillbirth Epidemiology investigator group. Stillbirths: rates, risk factors, and acceleration towards 2030. *Lancet* 2016; **387**: 587–603.
2. Lawn JE, Blencowe H, Oza S, You D, Lee AC, Waiswa P, Lalli M, Bhutta Z, Barros AJ, Christian P, Mathers C, Cousens SN; Lancet Every Newborn Study Group. Every Newborn: progress, priorities, and potential beyond survival. *Lancet* 2014; **384**: 189–205.
3. Lee ACC, Kozuki N, Blencowe H, Vos T, Bahalim A, Darmstadt GL, Niermeyer S, Ellis M, Robertson NJ, Cousens S, Lawn JE. Intrapartum-related neonatal encephalopathy incidence and impairment at regional and global levels for 2010 with trends from 1990. *Pediatr Res* 2013; **74** (Suppl 1): 50–72.
4. Bryant AS, Worjoloh A, Caughey AB, Washington AE. Racial/ethnic disparities in obstetric outcomes and care: prevalence and determinants. *Am J Obstet Gynecol* 2010; **202**: 335–343.
5. *Intrapartum Care: Care of Healthy Women and Their Babies During Childbirth* (Clinical Guideline 109). National Institute for Health and Care Excellence Clinical Guidelines 2014; www.nice.com.
6. Liston R, Sawchuck D, Young D; Society of Obstetrics and Gynaecologists of Canada; British Columbia Perinatal Health Program. Fetal health surveillance: antepartum and intrapartum consensus guideline. *J Obstet Gynaecol Can* 2007; **29** (Suppl 4): S3–S56.
7. Mangesi L, Hofmeyr GJ, Smith V, Smyth RM. Fetal movement counting for assessment of fetal wellbeing. *Cochrane Database Syst Rev* 2015; **10**: CD004909.
8. Alfirevic Z, Devane D, Gyte GM, Cuthbet A. Continuous cardiotocography (CTG) as a form of electronic fetal monitoring (EFM) for fetal assessment during labour. *Cochrane Database Syst Rev* 2017; **2**: CD006066.
9. Westergaard HB, Langhoff-Roos J, Lingman G, Marsál K, Kreiner S. A critical appraisal of the use of umbilical artery Doppler ultrasound in high-risk pregnancies: use of meta-analyses in evidence-based obstetrics. *Ultrasound Obstet Gynecol* 2001; **17**: 466–476.
10. Berkley E, Chauhan SP, Abuhamad A. Doppler assessment of the fetus with intrauterine growth restriction. *Am J Obstet Gynecol* 2012; **206**: 300–308.
11. Alfirevic Z, Stampalija T, Gyte GML, Neilson JP. Fetal and umbilical Doppler ultrasound in high-risk pregnancies. *Cochrane Database Syst Rev* 2017; **6**: CD007529.
12. Alfirevic Z, Stampalija T, Medley N. Fetal and umbilical Doppler ultrasound in normal pregnancy. *Cochrane Database Syst Rev* 2015; **4**: CD001450.
13. Baschat AA, Gembruch U. The cerebroplacental Doppler ratio revisited. *Ultrasound Obstet Gynecol* 2003; **21**: 124–127.
14. Twomey S, Flatley C, Kumar S. The association between a low cerebro-umbilical ratio at 30–34 weeks gestation, increased intrapartum operative intervention and adverse perinatal outcomes. *Eur J Obstet Gynecol Reprod Biol* 2016; **203**: 89–93.
15. Sabdia S, Greer RM, Prior T, Kumar S. Predicting intrapartum fetal compromise using the fetal cerebro-umbilical ratio. *Placenta* 2015; **36**: 594–598.
16. Khalil AA, Morales-Rosello J, Morlando M, Hannan H, Bhide A, Papageorghiou A, Thilaganathan B. Is fetal cerebroplacental ratio an independent predictor of intrapartum fetal compromise and neonatal unit admission? *Am J Obstet Gynecol* 2015; **213**: 54.e1–10.
17. Morales-Roselló J, Khalil A, Morlando M, Bhide A, Papageorghiou A, Thilaganathan B. Poor neonatal acid–base status in term fetuses with low cerebroplacental ratio. *Ultrasound Obstet Gynecol* 2015; **45**: 156–161.
18. Prior T, Mullins E, Bennett P, Kumar S. Prediction of intrapartum fetal compromise using the cerebroumbilical ratio: a prospective observational study. *Am J Obstet Gynecol* 2013; **208**: 124.e1–6.
19. Kowalczyk TD, Cabaniss M, Cusmano L. Association of low unconjugated estriol in the second trimester and adverse pregnancy outcome. *Obstet Gynecol* 1998; **91**: 396–400.
20. Schleifer RA, Bradley LA, Richards DS, Ponting NR. Pregnancy outcome for women with very low levels of maternal serum unconjugated estriol on second-trimester screening. *Am J Obstet Gynecol* 1995; **173**: 1152–1156.
21. Ay E, Kavak ZN, Elter K, Gokaslan H, Pekin T. Screening for pre-eclampsia by using maternal serum inhibin A, activin A, human chorionic gonadotropin, unconjugated estriol, and alpha-fetoprotein levels and uterine artery Doppler in the second trimester of pregnancy. *Aust N Z J Obstet Gynaecol* 2005; **45**: 283–288.
22. Schoen E, Norem C, O'Keefe J, Krieger R, Walton D, To TT. Maternal serum unconjugated estriol as a predictor for Smith–Lemli–Opitz syndrome and other fetal conditions. *Obstet Gynecol* 2003; **102**: 167–172.
23. Dugoff L, Hobbins JC, Malone FD, Vidaver J, Sullivan L, Canick JA, Lambert-Messerlian GM, Porter TF, Luthy DA, Comstock CH, Saade G, Eddleman K, Merkatz IR, Craigo SD, Timor-Tritsch IE, Carr SR, Wolfe HM, D'Alton ME; FASTER Trial Research Consortium. Quad screen as a predictor of adverse pregnancy outcome. *Obstet Gynecol* 2005; **106**: 260–267.

24. Chaiworapongsa T, Romero R, Korzeniewski SJ, Kusanovic JP, Soto E, Lam J, Dong Z, Than NG, Yeo L, Hernandez-Andrade E, Conde-Agudelo A, Hassan SS. Maternal plasma concentrations of angiogenic/antiangiogenic factors in the third trimester of pregnancy to identify the patient at risk for stillbirth at or near term and severe late preeclampsia. *Am J Obstet Gynecol* 2013; **208**: 287.e1–287.e15.
25. Crispi F, Llurba E, Domínguez C, Martín-Gallán P, Cabero L, Gratacós E. Predictive value of angiogenic factors and uterine artery Doppler for early- versus late-onset pre-eclampsia and intrauterine growth restriction. *Ultrasound Obstet Gynecol* 2008; **31**: 303–309.
26. Espinoza J, Romero R, Nien JK, Gomez R, Kusanovic JP, Gonçalves LF, Medina L, Edwin S, Hassan S, Carstens M, Gonzalez R. Identification of patients at risk for early onset and/or severe preeclampsia with the use of uterine artery Doppler velocimetry and placental growth factor. *Am J Obstet Gynecol* 2007; **196**: 326.e1–13.
27. Lobmaier SM, Figueras F, Mercade I, Perello M, Peguero A, Crovetto F, Ortiz JU, Crispi F, Gratacós E. Angiogenic factors *vs* Doppler surveillance in the prediction of adverse outcome among late-pregnancy small-for-gestational-age fetuses. *Ultrasound Obstet Gynecol* 2014; **43**: 533–540.
28. Yu J, Shixia CZ, Wu Y, Duan T. Inhibin A, activin A, placental growth factor and uterine artery Doppler pulsatility index in the prediction of pre-eclampsia. *Ultrasound Obstet Gynecol* 2011; **37**: 528–533.
29. Paiwattananupant K, Phupong V. Serum inhibin A level in preeclampsia and normotensive pregnancy. *Hypertens Pregnancy* 2008; **27**: 337–343.
30. Phupong V, Paiwattananupant K, Honsawek S. Inhibin A levels and severity of preeclampsia. *Arch Gynecol Obstet* 2009; **280**: 183–186.
31. Shen Z, Cai L-Y, Suprapto I-S, Shenoy P, Zhou X. Placental and maternal serum inhibin A in patients with preeclampsia and small-for-gestational-age. *J Obstet Gynaecol Res* 2011; **37**: 1290–1296.
32. Cemgil Arikan D, Ozkaya M, Adali E, Kilinc M, Coskun A, Ozer A, Bilge F. Plasma lipocalin-2 levels in pregnant women with pre-eclampsia, and their relation with severity of disease. *J Matern Fetal Neonatal Med* 2011; **24**: 291–296.
33. Heazell AEP, Whitworth M, Duley L, Thornton JG. Use of biochemical tests of placental function for improving pregnancy outcome. *Cochrane Database Syst Rev* 2015; **11**: CD011202.
34. Valiño N, Giunta G, Gallo DM, Akolekar R, Nicolaides KH. Biophysical and biochemical markers at 35–37 weeks' gestation in the prediction of adverse perinatal outcome. *Ultrasound Obstet Gynecol* 2016; **47**: 203–209.
35. Valiño N, Giunta G, Gallo DM, Akolekar R, Nicolaides KH. Biophysical and biochemical markers at 30–34 weeks' gestation in the prediction of adverse perinatal outcome. *Ultrasound Obstet Gynecol* 2016; **47**: 194–202.
36. Figueras F, Meler E, Iraola A, Eixarch E, Coll O, Figueras J, Francis A, Gratacós E, Gardosi J. Customized birthweight standards for a Spanish population. *Eur J Obstet Gynecol Reprod Biol* 2008; **136**: 20–24.
37. 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. *Am J Obstet Gynecol* 1985; **151**: 333–337.
38. Arduini D, Rizzo G. Normal values of pulsatility index from fetal vessels: a cross-sectional study on 1556 healthy fetuses. *J Perinat Med* 1990; **18**: 165–172.
39. Gómez O, Figueras F, Fernández S, Bennasar M, Martínez JM, Puerto B, Gratacós E. Reference ranges for uterine artery mean pulsatility index at 11–41 weeks of gestation. *Ultrasound Obstet Gynecol* 2008; **32**: 128–132.
40. Malin GL, Morris RK, Khan KS. Strength of association between umbilical cord pH and perinatal and long term outcomes: systematic review and meta-analysis. *BMJ* 2010; **340**: c1471.
41. Anderson NH, Sadler LC, Mckinlay CJ, Mccowan LME. INTERGROWTH 21st versus customized birthweight standards for identification of perinatal mortality and morbidity. *Am J Obstet Gynecol* 2015; **214**: 509.e1–e509.e7.
42. Bakalis S, Akolekar R, Gallo DM, Poon LC, Nicolaides KH. Umbilical and fetal middle cerebral artery Doppler at 30–34 weeks' gestation in the prediction of adverse perinatal outcome. *Ultrasound Obstet Gynecol* 2015; **45**: 409–420.
43. Akolekar R, Syngelaki A, Gallo DM, Poon LC, Nicolaides KH. Umbilical and fetal middle cerebral artery Doppler at 35–37 weeks' gestation in the prediction of adverse perinatal outcome. *Ultrasound Obstet Gynecol* 2015; **46**: 82–92.
44. Triunfo S, Crispi F, Gratacós E, Figueras F. Prediction of delivery of small-for-gestational-age neonates and adverse perinatal outcome by fetoplacental Doppler at 37 weeks' gestation. *Ultrasound Obstet Gynecol* 2017; **49**: 364–371.
45. Ilagan JG, Stamilio DM, Ural SH, Macones GA, Odibo AO. Abnormal multiple marker screens are associated with adverse perinatal outcomes in cases of intrauterine growth restriction. *Am J Obstet Gynecol* 2004; **191**: 1465–1469.
46. Gomez-Roig MD, Mazarico E, Sabria J, Parra J, Oton L, Vela A. Use of placental growth factor and uterine artery doppler pulsatility index in pregnancies involving intrauterine fetal growth restriction or preeclampsia to predict perinatal outcomes. *Gynecol Obstet Invest* 2015; **80**: 99–105.
47. Romero R, Nien JK, Espinoza J, Todem D, Fu W, Chung H, Kusanovic JP, Gotsch F, Erez O, Mazaki-Tovi S, Gomez R, Edwin S, Chaiworapongsa T, Levine RJ, Karumanchi SA. A longitudinal study of angiogenic (placental growth factor) and anti-angiogenic (soluble endoglin and soluble vascular endothelial growth factor receptor-1) factors in normal pregnancy and patients destined to develop preeclampsia and deliver a small for gestational age neonate. *J Matern Fetal Neonatal Med* 2008; **21**: 9–23.
48. Neilson JP. Biochemical tests of placental function for assessment in pregnancy. *Cochrane Database Syst Rev* 2012; **8**: CD000108.
49. Benton SJ, McCowan LM, Heazell AEP, Grynspan D, Hutcheon JA, Senger C, Burke O, Chan Y, Harding JE, Yockell-Lelièvre J, Hu Y, Chappell LC, Griffin MJ, Shennan AH, Magee LA, Gruslin A, von Dadelszen P. Placental growth factor as a marker of fetal growth restriction caused by placental dysfunction. *Placenta* 2016; **42**: 1–8.
50. 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 Obstet Gynecol* 2000; **15**: 209–212.
51. Jain M, Farooq T, Shukla RC. Doppler cerebroplacental ratio for the prediction of adverse perinatal outcome. *Int J Gynaecol Obstet* 2004; **86**: 384–385.
52. 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. *J Ultrasound Med* 2005; **24**: 1223–1228.
53. Bahado-Singh RO, Kovanci E, Jeffres A, Oz U, Deren O, Copel J, Mari G. The Doppler cerebroplacental ratio and perinatal outcome in intrauterine growth restriction. *Am J Obstet Gynecol* 1999; **180**: 750–756.
54. Severi FM, Bocchi C, Visentin A, Falco P, Cobellis L, Florio P, Zagonari S, Pilu G. Uterine and fetal cerebral Doppler predict the outcome of third-trimester small-for-gestational age fetuses with normal umbilical artery Doppler. *Ultrasound Obstet Gynecol* 2002; **19**: 225–228.
55. Cruz-Martínez R, Figueras F, Hernandez-Andrade E, Oros D, Gratacós E. Fetal brain Doppler to predict cesarean delivery for nonreassuring fetal status in term small-for-gestational-age fetuses. *Obstet Gynecol* 2011; **117**: 618–626.
56. Figueras F, Gratacós E. Stage-based approach to the management of fetal growth restriction. *Prenat Diagn* 2014; **34**: 655–659.
57. Figueras F, Gratacós E. Update on the diagnosis and classification of fetal growth restriction and proposal of a stage-based management protocol. *Fetal Diagn Ther* 2014; **36**: 86–98.

SUPPORTING INFORMATION ON THE INTERNET

The following supporting information may be found in the online version of this article:

Table S1 Univariate regression analysis to determine significant contributors to prediction of adverse perinatal outcome (APO) in the general population and in small-for-gestational-age (SGA) fetuses