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A prediction model for short-term neonatal outcomes in severe early-onset fetal growth restriction

Andrew Sharp[a,*], Richard Jackson[b], Christine Cornforth[b], Jane Harrold[b], Mark A. Turner[a], Louise Kenny[a], Philip N. Baker[c], Edward D. Johnstone[d], Asma Khalil[e,f], Peter von Dadelszen[g], Aris T. Papageorghiou[e,f], Zarko Alfirevic[a]

[a] Department of Women's and Children's Health, University of Liverpool and The Liverpool Women's Hospital, Members of Liverpool Health Partners, United Kingdom
[b] Liverpool Clinical Trials Unit, University of Liverpool, United Kingdom
[c] College of Life Sciences, University of Leicester, United Kingdom
[d] Maternal & Fetal Health Research Centre, School of Medical Sciences, Faculty of Medicine Biology and Health, University of Manchester, United Kingdom
[e] Fetal Medicine Unit, St George's Hospital, University of London, United Kingdom
[f] Vascular Biology Research Centre, Molecular and Clinical Sciences Research Institute, St George's, University of London, Cranmer Terrace, London SW17 0RE, United Kingdom
[g] Department of Women's and Children's Health, School of Life Course Sciences, King's College London, United Kingdom

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ABSTRACT

Background: Severe early-onset fetal growth restriction (FGR) predisposes to fetal death, neonatal death, neonatal morbidity and neurodisability. The use of placental biomarkers has been proposed for risk stratification in pre-eclampsia, but they could be equally useful in fetal growth restriction in aiding management.

Objective: To determine the efficacy of angiogenic biomarkers at predicting adverse pregnancy outcome in severe early-onset fetal growth restriction.

Study design: This is a secondary analysis of the multicentre, placebo-controlled STRIDER UK randomised controlled trial of singleton pregnancies with severe early-onset fetal growth restriction.

Women with FGR pregnancies between 22^{+0} and 29^{+6} weeks of gestation were randomly assigned to receive either sildenafil 25 mg three times daily or placebo until 32^{+0} weeks' gestation or delivery. We developed prediction models based upon maternal demographics (age, parity, blood pressure, preeclampsia, gestational hypertension), fetal biometric (estimated fetal weight) and Doppler measurements (Middle Cerebral Artery (MCA), Umbilical Artery (UA)) and maternal angiogenic biomarkers [placental growth factor (PlGF), soluble endoglin (sEng), soluble fms-like tyrosine kinase 1 (sFlt-1) and sFlt-1:PlGF ratio) using both univariate and multivariate analysis.

Results: A complete data set was available for 105 of 135 randomised women. Multivariate regression analysis identified estimated fetal weight (EFW) and sFlt-1:PlGF as independent predictors of livebirth (EFW OR: 1.01 (1.008, 1.021); $p < 0.001$ and lower sFlt-1:PlGF ratio OR: 0.53 (0.284, 0.994); $p = 0.048$) and overall survival (EFW OR: 1.01 (1.006, 1.015); $p < 0.001$ and lower sFlt-1/PlGF ratio OR: 0.51 (0.286, 0.904); $p = 0.021$). EFW was a consistent predictor for all outcomes other than gestation at delivery. sFlt-1:PlGF ratio was a consistent predictor for all outcomes other than neonatal morbidity.

Conclusions: In severe early-onset FGR pregnancies livebirth and overall survival can be predicted using a model involving EFW and sFlt-1:PlGF ratio. This model require validation in a larger cohort but may allow informed decision making about pregnancy management, especially in previable cases.

Introduction

Severe early-onset fetal growth restriction (FGR) is associated with significant adverse pregnancy outcomes; fetal and neonatal death [1–3], necrotizing enterocolitis [3], respiratory complications [3], neurodisability [4–7] and lifelong health risks for the

* Corresponding author at: Department of Women's and Children's Health Research, University of Liverpool, Liverpool Women's Hospital, Crown Street, Liverpool, L8 7SS, United Kingdom.
E-mail address: asharp@liv.ac.uk (A. Sharp).

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child [8–10]. Currently, there is no effective treatment for severe early-onset FGR with women being offered a choice of; 1) expectant management with intensive surveillance and iatrogenic preterm delivery or 2) termination of pregnancy if available [11].

We conducted a randomised controlled trial (RCT) to test the hypothesis that sildenafil, a phosphodiesterase 5 (PDE5) inhibitor, could prolong gestation by improving the blood supply to the placental bed [12]. However, our RCT demonstrated no evidence of benefit in either short-term fetal or neonatal outcomes in those women treated with sildenafil [13]. Despite these negative findings, the STRIDER UK RCT does provide valuable clinical and biomarker information for early-onset FGR pregnancies with a more extreme phenotype than other previously published cohorts [1,11].

Risk stratification on which to base a prognosis for the pregnancy, such as the likelihood of the fetus being born alive or surviving the neonatal period is currently lacking. This may be particularly pertinent when the diagnosis of FGR is made at extremely early gestations when viability is uncertain.

Published FGR cohorts such as the TRUFFLE study provide data on the risk of perinatal mortality and neurological impairment at 2 years of age in pregnancies with moderate to severe FGR [7,11]. The TRUFFLE study demonstrated that the risk of fetal demise is actually very low (<2%) in fetuses with an estimated fetal weight (EFW) is >500 g and an abnormal umbilical artery Doppler (raised PI or worse) when management is led by a fetal medicine expert guided by ultrasound and computerized CTG. Another large cohort of severe early-onset FGR fetuses, classified as previable, showed a perinatal mortality of 52% (111/212, after excluding terminations) and a diagnosis to delivery interval of 8.1 weeks for survivors [1]. Unfortunately, neither of these studies had information on placental biomarkers.

With regards to prediction of FGR, the most effective biomarker to date appears to be placental growth factor (PlGF) [14–17]. A panel of angiogenic biomarkers, including PlGF and soluble fms-like tyrosine kinase 1 (sFlt-1), measured at 24–28 weeks of gestation appears to be a good predictor of subsequent fetal demise (Relative risk of 29.1) when grossly abnormal [18] a feature that persists at later gestations [19]. A recent large cohort study investigating the use of routine third trimester ultrasound screening had a minimal impact on stillbirth [20] but when combined with sFlt-1/PlGF would have reliably predicted FGR at 28 weeks (Positive Likelihood Ratio [LR+] 41.1, Sensitivity 38.5%, Specificity 99.1% and Positive Predictive Value [PPV] 21.3) and predicted delivery of an FGR baby with associated preeclampsia or perinatal morbidity when performed at 36 weeks (LR + 17.5, Sensitivity 37.9%, Specificity 97.8% and PPV 21.6) [21]. Small cohort studies in early onset FGR populations have confirmed this association with abnormal sFlt-1/PlGF ratio [22,23].

In light of the promising data from small studies on biomarker prediction [22,23] and the significant pathology in our STRIDER UK cohort, we hypothesized that a prediction model based upon measurable clinical features on ultrasound and biomarkers for placental disease in a population of fetuses with severe early-onset FGR could be beneficial to aid decision making by women and clinicians.

Materials and methods

This is a secondary analysis of the association between fetal biometric measurements, Doppler indices and maternal angiogenic biomarkers at the time of diagnosis of severe early-onset FGR with pregnancy outcome from the STRIDER UK trial [13].

STRIDER UK is a multicentre RCT of sildenafil vs placebo for the treatment of severe early-onset FGR defined as a singleton pregnancy between 22^{+0} and 29^{+6} weeks of gestation with; i) a fetus with an abdominal circumference (AC) or EFW below the 10^{th} centile and ii) absent or reversed end diastolic flow (EDF) in the umbilical artery on Doppler velocimetry. Gestation was determined by first trimester crown rump length.

Following informed consent and biometry assessment women were randomised to receive sildenafil or placebo (25 mg three times per day) until 32^{+0} weeks of gestation or delivery, with the clinical management and decision to deliver determined by the attending clinical team. Doppler, growth and blood pressure were assessed a minimum of weekly by a fetal medicine specialist. Blood samples were taken prior to treatment and at regular points over the following 2 weeks after randomization. There was no change in angiogenic blood parameters over time as demonstrated by the fitting of longitudinal models and assessing the slope term (data not shown). As no change was observed, only blood biomarkers from the time of diagnosis of FGR have been considered for this analysis. The use of Sildenafil was also included in these models and the change in angiogenic markers showed no difference between treatment arms.

Doppler ultrasound was performed serially in four vessels; the umbilical artery (UA), middle cerebral artery (MCA), ductus venosus (DV) and uterine artery (UtA). In addition to the Pulsatility Index (PI), UA EDF, DV a-wave and the presence of bilateral UtA notching were recorded. Abnormal Doppler findings were defined as follows; for UA raised PI (>95^{th} centile), absent EDF, or reversed EDF; for MCA low PI (<5^{th} centile); for DV a-wave absent or reversed, and for UtA mean PI > 1.45 or bilateral notching.

Serum samples (≥2 ml) collected at the time of diagnosis of FGR and prior to treatment were analyzed retrospectively. Maternal serum concentrations of sFlt-1 and PlGF (pg/ml) were determined using the automated Elecsys® electro-chemiluminescence immunoassay platform (Roche Cobas, Mannheim, Germany) and used to calculate sFlt-1:PlGF ratios. Maternal serum concentrations of soluble endoglin (sEng) (ng/ml) were determined using human Quantikine® enzyme-linked immunosorbent assays (R&D Systems, Minneapolis, MN, USA).

Statistical methodology

Univariate and Multivariable generalized linear models were used to assess the impact of clinical covariates and biomarkers on neonatal outcomes. All continuous clinical covariates at randomization and categorical clinical covariates with sufficient frequencies to be discriminatory (at least 5 observations in each category) were included in the univariate analysis; gestational age, gestational hypertension, preeclampsia, EFW, blood pressure (systolic and diastolic), mean arterial pressure (MAP) and parity. Doppler covariates included UA EDF, DV a-wave, MCA PI and UtA notching. Biomarker data included PlGF, sFlt-1, sEng and sFlt-1:PlGF ratio. EFW was included in the model as it was observed with effects relative to each 100 g. Models were explored that included gestational age with EFW, gestational age alone, and EFW corrected for gestational age (e.g. As a Z-score or the expected EFW for the gestational age). However, the best model performance was for the model which included EFW alone. Gestational age was measured in days and then converted into weeks for clarity of presentation. No precision or granularity was lost in this conversion.

Analyses were carried out on six clinical outcomes; livebirth, gestation at delivery, overall survival, neonatal morbidity, birth weight and a composite measure of overall survival or neonatal morbidity. Overall survival was defined as a hospital discharge of a live child. Neonatal morbidity was defined as a liveborn fetus surviving to discharge and experiencing at least one of the following adverse outcomes: necrotizing enterocolitis, retinopathy of prematurity, bronchopulmonary dysplasia requiring oxygen therapy at 36 weeks, patent ductus arteriosus requiring medical or

surgical treatment, the need for vasopressor therapy, neonatal infection, intraventricular hemorrhage (IVH) within 6 weeks of delivery or a confirmed serious adverse event (SAE) as defined by the STRIDER UK protocol [13].

Univariate analyses and multivariate analyses were carried out using a generalized linear modelling approach assuming a Gaussian family with identity link for continuous outcomes and a binomial family with logistic link for categorical outcomes. All terms included were considered as candidate covariates in the multivariate analysis. Selection of terms in multivariate models were performed using a forward stepwise approach evaluated using Akaikes Information Criterion (AIC). Continuous covariates were evaluated graphically and models produced which included both raw and transformed data. The models themselves were constructed using a forwards stepwise procedure with other 'plausible alternatives' also explored by adding and removing terms and assessing model parameters to assess if any multi-colinearity was having any adverse effect on the models. A forward approach was chosen over a backwards approach to avoid over-parameterized models which would include a large number of candidate covariates. Values of PlGF, sFlt-1, sEng, and sFlt-1:PlGF ratio were included as covariates measured on the log scale to account for extreme value observations. The distribution of all angiogenic markers were assessed following transformation to ensure they were appropriately distributed for further analysis. Reported results were presented in terms of odds ratios (95% CI) for categorical data and mean (95% CI) estimates for continuous outcomes. Interval validation of the final model for each outcome was carried out using a bootstrap approach using measures of discrimination and calibration. AUCs between models with and without biomarkers were compared with De Longs test. Graphical summaries present the predicted model results along with all analyses are carried out using the statistical package R (Version 3).

Results

Study population

One hundred and thirty five women were recruited to the STRIDER UK trial between November 2014 and July 2016 from 19 fetal medicine units within the UK. The study population available for analysis was 105 (77.8%) women (Table 1); data for 30 of the recruited women was removed as angiogenic biomarker information was unavailable. Sixty one (58%) women were recruited before 26^{+0} weeks and 44 (42%) between 26^{+0} and 29^{+6} weeks of gestation. Of the 105 available participants, 70/105 (67%) babies were born alive and 59/105 (56%) were discharged alive. 46/70 (66%) liveborn babies experienced neonatal morbidity. The median (IQR) gestation at delivery was 28.3 (26.9, 29.7) weeks and the median (IQR) birth weight was 590 g (480, 769).

Modelling

The results of univariate analysis for each of the five outcomes are included in Table 2. Considering clinical covariates, EFW at randomization (considered synonymous with confirmation of diagnosis of FGR) and gestation at randomization were consistent univariate predictors for all outcomes. Preeclampsia was a univariate indicator of a lower gestational age at delivery (est [se] = −1.06 [0.506]; p-value = 0.038). Reversed UA EDF Doppler (est [se] = −1.08 [0.417]; p-value = 0.011) was a univariate indicator of gestation at delivery. All four biomarker measures (PlGF, sEng, sFlt-1 and sFlt-1:PlGF ratio) were univariate indicators of gestation at delivery and birth weight. UtA Doppler was also a significant indicator for birth weight. PlGF and the sFlt-1:PlGF ratio were significant univariate indicators of livebirth and overall survival.

Table 1
Demographic data for the STRIDER UK study cohort.

Covariate	Level	Total n = 105
Age	Median (IQR)	30 (27, 35)
Previous Pregnancy	No	54
	Yes	51
Gestational Hypertension	No	81
	Yes	24
Pre-Eclampsia	No	87
	Yes	18
Diastolic Blood Pressure	Median (IQR)	88.5 (79.5, 95.5)
Systolic Blood Pressure	Median (IQR)	134.5 (124.5, 144.5)
Mean Arterial Pressure	Median (IQR)	104.2 (93.5, 111.7)
Estimated Fetal Weight	Median (IQR)	451 (369, 651)
Gestation at Randomization	Median (IQR)	25.3 (24.1, 27.4)
UA EDF	Absent	74
	Reversed	29
DV a-wave	A wave Positive	93
	A wave Reversed	7
MCA	Abnormal	53
	Normal	42
UtA	No Notch (<1.45)	13
	No Notch (≥1.45)	7
	Notch	73
Birth Status	Stillbirth	35
	Livebirth	70
Gestation at Delivery	Median (IQR)	28.3 (26.9, 29.7)
Neonatal Outcome	Died (SB/NND)	46
	Survived	59
Neonatal Morbidity	No	24
	Yes	46
Birthweight	Median (IQR)	590 (480, 769)
PlGF pg/ml	Median (IQR)	22 (13, 42)
sEng ng/ml	Median (IQR)	33.6 (20.4, 57.8)
sFlt-1pg/ml	Median (IQR)	6894 (5025, 10,380)
sFlt-1/PlGF pg/ml	Median (IQR)	343.9 (167.2, 591.6)

The results of the selected univariate modelling are included in Table 2. The results of multivariate modelling are included in Table 3 and include model intercepts for completeness. The results for each outcome are discussed separately.

Livebirths

As EFW increased, the odds of a live birth also increased [OR: 4.28 (2.299, 7.975); p-value < 0.001] per 100 g of EFW (Fig. 1). A lower sFlt-1:PlGF ratio [OR: 0.53 (0.284, 0.994); p-value = 0.048] was associated with a larger probability of overall neonatal survival. In terms of measuring the importance of each covariate, EFW can be shown to explain 40% of the model variability whereas sFlt-1:PlGF ratio explains 4%. The intercept of 0.14 (0.003, 7.39) allows estimation of the probability of a live birth (Fig. 2A, B). This data is also represented as a receiver operating characteristic (ROC) curve with an area under the curve (AUC) of 0.90 (Fig. 2C). For example; for a fetus with an estimated fetal weight of 400 g and a (log) sFlt-1:PlGF ratio of 4, the estimated probability of a live birth would be 37% as shown below.

$$logit(p) = \log(0.14) + 400\log(1.01) + 4\log(0.53)$$

$$p = \ 0.3716$$

Gestation at birth

The presence of pre-eclampsia reduced the gestation at birth by almost a week [Est: −0.97 (−1.8, −0.2); p-value = 0.020] whilst having a later gestational age at randomization delayed the gestation at birth [Est: 0.61 (0.5, 0.7); p-value < 0.001]. Regarding Doppler measurement, the presence of reversed EDF in the umbilical artery [Est: −0.97 (−1.6, −0.3); p-value < 0.001] resulted in an earlier gestation at delivery. With respect to biomarker data, a

Table 2
Univariate analysis for five clinical outcomes (Livebirth, Gestation at Birth (weeks), Overall Survival, Neonatal Morbidity and Birth Weight (grams).

Covariate	Level	Livebirth		Gestation at Birth (weeks)		Overall Survival		Neonatal Morbidity		Birth Weight (grams)		Composite Outcome[a]	
		Est	P	Est	P	Est	P	Est	P	Est	P	Est	P
Allocation	Placebo												
	Sildenafil	1.19 (0.578, 2.464)	0.632	−0.42 (0.392)	0.285	1.01 (0.514, 1.999)	0.969	1.81 (0.75, 4.369)	0.187	−13.41 (56.726)	0.813	1.28 (0.626, 2.606)	0.501
Age		0.97 (0.911, 1.039)	0.411	−0.02 (0.035)	0.615	0.98 (0.925, 1.045)	0.587	1.01 (0.932, 1.089)	0.852	0.28 (5.101)	0.956	0.97 (0.912, 1.038)	0.404
Previous pregnancy	No												
	Yes	1.02 (0.49, 2.105)	0.967	0.29 (0.395)	0.471	0.85 (0.43, 1.684)	0.642	1.32 (0.544, 3.193)	0.54	71.91 (56.71)	0.207	1.06 (0.518, 2.174)	0.871
Gestational Hypertension	No												
	Yes	1.23 (0.53, 2.862)	0.629	−0.22 (0.449)	0.623	0.93 (0.431, 2.022)	0.861	0.64 (0.245, 1.661)	0.357	1.56 (64.691)	0.981	0.99 (0.439, 2.223)	0.976
Pre-Eclampsia	No												
	Yes	1.5 (0.549, 4.097)	0.429	−1.06 (0.506)	0.038	0.93 (0.385, 2.266)	0.88	2.87 (0.76, 10.827)	0.119	−73.44 (73.876)	0.322	1.32 (0.502, 3.445)	0.577
Diastolic Blood Pressure		1.01 (0.984, 1.032)	0.532	0.01 (0.005)	0.142	1.01 (0.988, 1.025)	0.471	0.98 (0.942, 1.019)	0.298	1.46 (0.699)	0.039	1.01 (0.988, 1.022)	0.552
Systolic Blood Pressure		1 (0.973, 1.019)	0.711	−0.02 (0.012)	0.079	0.99 (0.969, 1.012)	0.394	1 (0.972, 1.033)	0.897	−1.54 (1.801)	0.395	0.99 (0.97, 1.015)	0.515
Mean Arterial Pressure		1.01 (0.987, 1.026)	0.54	0.01 (0.007)	0.288	1.01 (0.99, 1.021)	0.513	0.99 (0.958, 1.013)	0.299	1.81 (1.00)	0.072	1 (0.988, 1.021)	0.613
Estimated Fetal Weight		4.75 (2.686, 8.402)	<0.001	0.68 (0.069)	<0.001	3.09 (2.1, 4.557)	<0.001	0.69 (0.543, 0.875)	0.002	1.31 (0.066)	<0.001	4 (2.424, 6.594)	<0.001
Gestation at Randomisation		2.16 (1.623, 2.874)	<0.001	0.68 (0.067)	<0.001	2 (1.566, 2.542)	<0.001	0.63 (0.485, 0.824)	0.001	103.23 (9.276)	<0.001	1.98 (1.533, 2.564)	<0.001
EDF	Absent												
	Reversed	0.8 (0.37, 1.732)	0.571	−1.08 (0.417)	0.011	0.65 (0.313, 1.359)	0.254	1.64 (0.603, 4.475)	0.331	−57.29 (61.465)	0.353	0.8 (0.373, 1.71)	0.564
MCA	Abnormal												
	Normal	1.42 (0.653, 3.081)	0.377	0.56 (0.427)	0.192	1.05 (0.511, 2.148)	0.9	0.93 (0.372, 2.341)	0.883	28.44 (61.676)	0.646	1.19 (0.555, 2.538)	0.658
UA	No Notch (<1.45)												
	No Notch (≥1.45)	0.43 (0.08, 2.308)	0.324	0.84 (0.138, 5.07)	0.847	0.64 (0.134, 3.029)	0.57	0.28 (0.037, 2.092)	0.214	−16.7 (129.571)	0.898	0.43 (0.08, 2.308)	0.324
	Notch	0.48 (0.146, 1.574)	0.225	0.33 (0.102, 1.079)	0.069	0.62 (0.222, 1.749)	0.368	1.42 (0.413, 4.902)	0.576	−181.21 (84.746)	0.035	0.44 (0.133, 1.432)	0.171
PlGF		3.89 (1.9, 7.956)	<0.001	1.4 (0.219)	<0.001	3.67 (1.903, 7.058)	<0.001	0.59 (0.336, 1.04)	0.068	262.6 (28.438)	<0.001	3.75 (1.858, 7.555)	<0.001
sEng		0.77 (0.455, 1.303)	0.331	−0.67 (0.274)	0.015	0.63 (0.381, 1.045)	0.073	1.22 (0.672, 2.227)	0.509	−115.58 (40.862)	0.006	0.67 (0.398, 1.143)	0.144
sFlt-1		1.12 (0.611, 2.036)	0.722	−0.95 (0.329)	0.005	0.88 (0.49, 1.564)	0.653	1.53 (0.769, 3.046)	0.226	−124.97 (49.403)	0.013	1.05 (0.576, 1.91)	0.877
sFlt-1/PlGF		0.56 (0.355, 0.872)	0.011	−0.99 (0.157)	<0.001	0.51 (0.327, 0.782)	0.002	1.42 (0.978, 2.075)	0.065	−169.72 (21.821)	<0.001	0.55 (0.348, 0.857)	0.008

[a] Composite outcome of Overall Survival and Neonatal Morbidity.

Table 3
Multivariate analysis for five clinical outcomes (Livebirth, Gestation at Birth (weeks), Overall Survival, Neonatal Morbidity and Birth Weight (grams).

Covariate	Level	Livebirth		Gestation at Birth (weeks)		Overall Survival		Neonatal Morbidity		Birth Weight (grams)		Composite Outcome[b]	
		OR (95% CI)	P	Est (95% CI)	P	OR (95% CI)	P	OR (95% CI)	P	Est (95% CI)	P	OR (95% CI)	P
Intercept		0.14 (0.003, 7.39)	0.33	16.67 (12.5, 20.9)	<0.001	0.43 (0.013, 14.798)	0.641	21.72 (4.275, 110.3)	<0.001	1050.38 (544.26, 1556.51)	<0.001	0.43 (0.013, 14.798)	0.641
Pre-Eclampsia	No												
	Yes			−0.97 (−1.8, −0.2)	0.02								
EDF	Absent												
	Reversed			−0.97 (−1.6, −0.3)	<0.001								
Estimated Fetal Weight	median (IQR)	4.28 (2.299, 7.975)	<0.001			2.86 (1.845, 4.438)	<0.001	0.69 (0.543, 0.875)	0.002	138 (114, 161)	<0.001	3.91 (2.186, 6.997)	<0.001
Previous Pregnancy	Yes										0.012		
	No									66.36 (15.67, 117.05)	0.026		
Gestation at Randomisation	median (IQR)			0.61 (0.5, 0.7)	<0.001					−26.62 (−49.64, −3.61)	0.015		
sEng	median (IQR)									61.94 (12.72, 111.15)	<0.001		
sFlt-1/PlGF	median (IQR)	0.53 (0.284, 0.994)	0.048	−0.6 (−0.8, −0.3)	<0.001	0.51 (0.286, 0.904)	0.021			−119.43 (−151.94, 86.93)		0.53 (0.284, 0.976)	0.042
Model Performance	**AUC**	**90.40%** (84.8%–95.6%)		**90.7%**[a] (83.9 –97.2%)		**88.40%** (82.5%– 94.4%)		**70.30%** (58.2%–80.7%)		**94.48%**[a] (90.5% –97.9%)		**89.60%** (84.0% –95.5%)	

[a] AUC calculated based on weighted average of AUCs using each unique covariate value to dichotomize into a binary covariate.
[b] Composite outcome of Overall Survival and Neonatal Morbidity.

higher sFlt-1:PlGF ratio [Est: −0.6 (−0.8, −0.3); p-value < 0.001] led to an earlier gestation at delivery.

Overall survival

As EFW increased, the odds of overall survival increased [OR: 2.86 (1.845, 4.438) p-value < 0.001]. A lower sFlt-1:PlGF ratio [OR: 0.51 (0.286, 0.904); p-value = 0.021] was also associated with a larger probability of overall survival. Fig. 3 shows the relationship between EFW and sFlt-1:PlGF ratio at diagnosis and survival. A graphical representation of the model results is also provided to give predicted probabilities of overall survival based on EFW and sFlt-1:PlGF ratio (Fig. 3A, B). This data is also represented as a ROC curve with an AUC of 0.88 (Fig. 3C).

Neonatal morbidity

The only covariate chosen for inclusion in the multivariate model of neonatal morbidity was EFW. As EFW increased the probability of neonatal morbidity decreased [Est: 0.69 (0.543, 0.875) p-value = 0.002].

Birth weight

Birth weight was greater if the EFW [Est: 138 (114, 161); p-value < 0.001] at first scan was larger. However, if the gestation at first scan was higher there was a negative impact on birth weight [Est: −26.62 (−49.64, −3.61); p-value = 0.026]. Considering clinical characteristics, primiparity was associated with a greater birth weight [Est: 66.36 (15.67, 117.05); p-value = 0.012]. Considering biomarker covariates, an increased level of sEng [Est: 61.94 (12.72, 111.15); p-value = 0.015] was associated with greater birth weight whereas an increased sFlt-1:PlGF ratio [Est: −119.43 (−151.94, 86.93); p-value < 0.001] was associated with lower birth weight.

Composite overall survival and neonatal morbidity

As EFW increased, the odds of an adverse outcomes increased [OR: 3.91 (2.186, 6.997); p-value < 0.001]. A lower sFlt-1:PlGF ratio [OR: 0.53 (0.284, 0.976); p-value = 0.042] was also associated with a larger probability of an adverse outcome.

For the models predicting birth status, overall survival and neonatal morbidity EFW is included as a predictive covariate as measured (e.g. not adjusting for gestational age). Models were explored that included gestational age with EFW, gestational age alone, and EFW corrected for gestational age (e.g. As a Z-score or the expected EFW for the gestational age). However, the best model performance was for the model that included EFW, so other ways to express fetal weight are not included in the reported models. However, recognizing that gestational age is an important predictor of long term neonatal outcome, model investigations were performed replacing EFW at randomization with gestation at randomization. EFW and gestation are highly correlated (Pearson's correlation = 0.87). A comparison of the models are given in the Supplementary information (Table S1). This demonstrates that a model including gestational age at randomization has both a larger model deviance (104.9 compared to 89.0) and a smaller measure of concordance (c-statistic) (0.88 compared to 0.83) and thus the model including EFW rather than gestational age at randomisation has a better model fit. To further illustrate the correlation of EFW and gestational age at randomisation, we have added Fig. 1 as a graphical representation of a model including EFW and gestational age to predict Overall Survival without biomarkers.

The impact of the sFlt-1:PlGF ratio did not materially change between the two models. A further summary of the internal validation used to assess the performance of each model is included in the Supplementary information (Table S2).

For each response, comparisons were made between models both with and without biomarkers. Comparisons of neonatal Morbidity are not included, as the final model does not include any

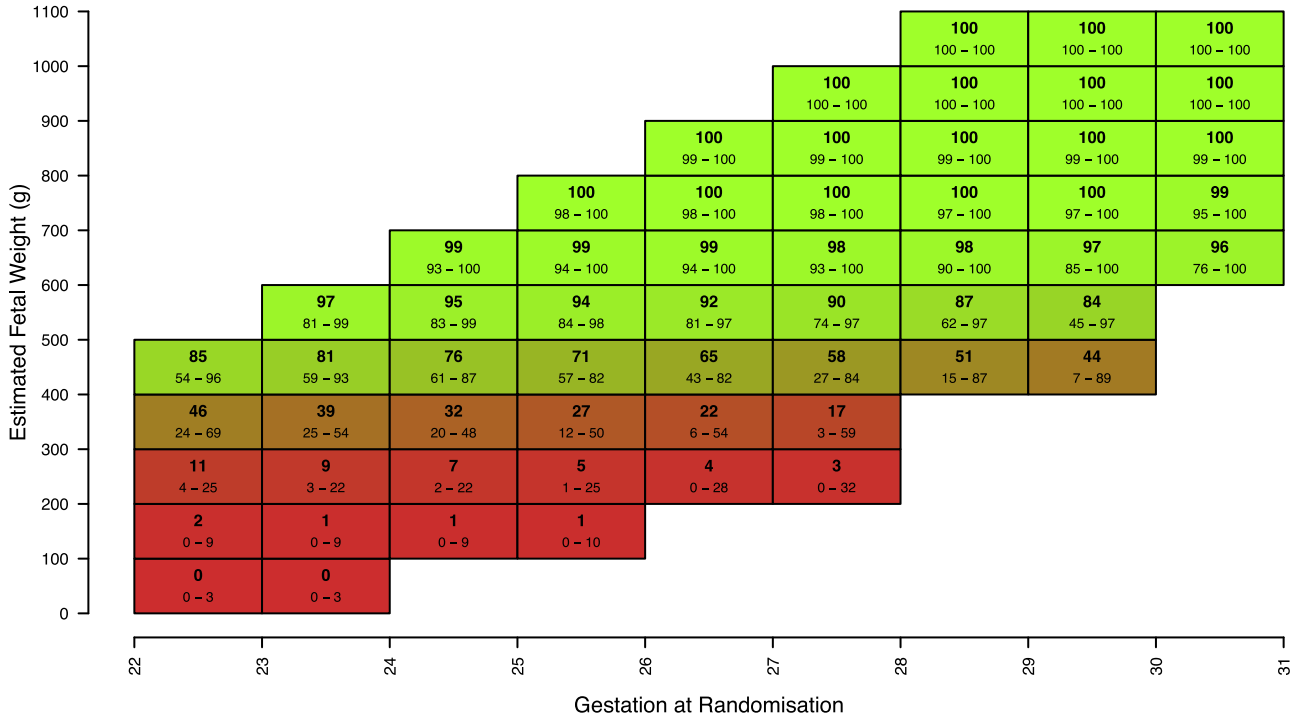

Fig. 1. Overall survival by estimated fetal weight and gestation at diagnosis of FGR.

biomarker covariates. The additional predictive potential of biomarkers are compared using De Long's test and whilst none of the model suggest that the inclusions of biomarkers are significant at the 5% level, this may be explained by the high performance of models without biomarkers and the relatively small datasets on which models are constructed. Estimation of Overall Survival (from 85.7% to 88.4%) and Birth weight (from 91.7% to 94.5%) show the largest improvements. Full details are in the Supplementary information (Table S3).

Discussion

Principal findings

Our study has confirmed that the combination of clinical biometric data routinely recorded in cases of early-onset FGR and sFlt-1:PlGF ratio can predict pregnancy outcome for both live-birth, gestation at delivery, birthweight and overall survival. Other covariates also showed benefit in predicting gestation at delivery and birth weight. EFW was a consistent predictor for all outcomes other than gestation at delivery. sFlt-1:PlGF ratio was a consistent predictor for all outcomes other than neonatal morbidity.

Results

To date the majority of published data have demonstrated that a low PlGF or raised sFlt-1:PlGF ratio are associated with a greater likelihood of stillbirth [16,24–27] or adverse pregnancy outcome [28] and is associated with fetal growth restriction [2,17,21,29–33] and placental pathology [16]. A recent Cochrane Diagnostic Test Accuracy Review on the effectiveness of biomarkers to predict stillbirth calculated that an abnormal PlGF or sFlt-1:PlGF ratio have a diagnostic odds ratio of 49.2 for subsequent stillbirth [34]. Our data appears to corroborate the accuracy of sFlt-1:PlGF ratio for predicting adverse pregnancy outcome with livebirth and overall survival having an AUC of 0.90 and 0.88 respectively.

This finding is logical as a poorly functioning placenta would be expected to have associated angiogenic factor dysfunction as observed in preeclampsia [17,27]. The fact that in pregnancies affected by severe early-onset FGR this finding can be used to estimate pregnancy outcome is, however, novel.

Clinical implications

The strength of our model is that it combines the diagnostic sensitivity of the sFlt-1:PlGF ratio with the routinely collected clinical covariates of EFW and gestational age to determine likelihood of livebirth and overall survival. Whilst the actual improvement in prediction from sFlt-1/PlGF ratio over gestational age and EFW may be small its clinical implications may be substantial.

The results of this study cannot readily be generalized beyond our inclusion criteria to other high risk pregnancy situations or late-onset FGR, and would need to be further validated within an FGR population. However, within the context of severe early-onset FGR we feel that providing a predictive assessment of outcome will be of significant value to clinicians and parents

Research implications

Several small cohort studies have shown an association between early onset FGR and abnormal sFlt-1/PlGF ratio [22,23]. A recent large prospective cohort was also able to use this biomarker to predict subsequent delivery with a FGR fetus but in a low risk unselected population [21]. Due to the close association between pregnancy outcome and PlGF or sFlt-1:PlGF ratio it is perhaps unsurprising that we identified no effect from the use of the angiogenic markers sEng or sFlt-1 alone. Further research would potentially clarify the role of these biomarkers in pregnancy prediction.

Strengths and limitations

This study is not the first to study the predictive capacity of angiogenic biomarkers to predict adverse pregnancy outcome in

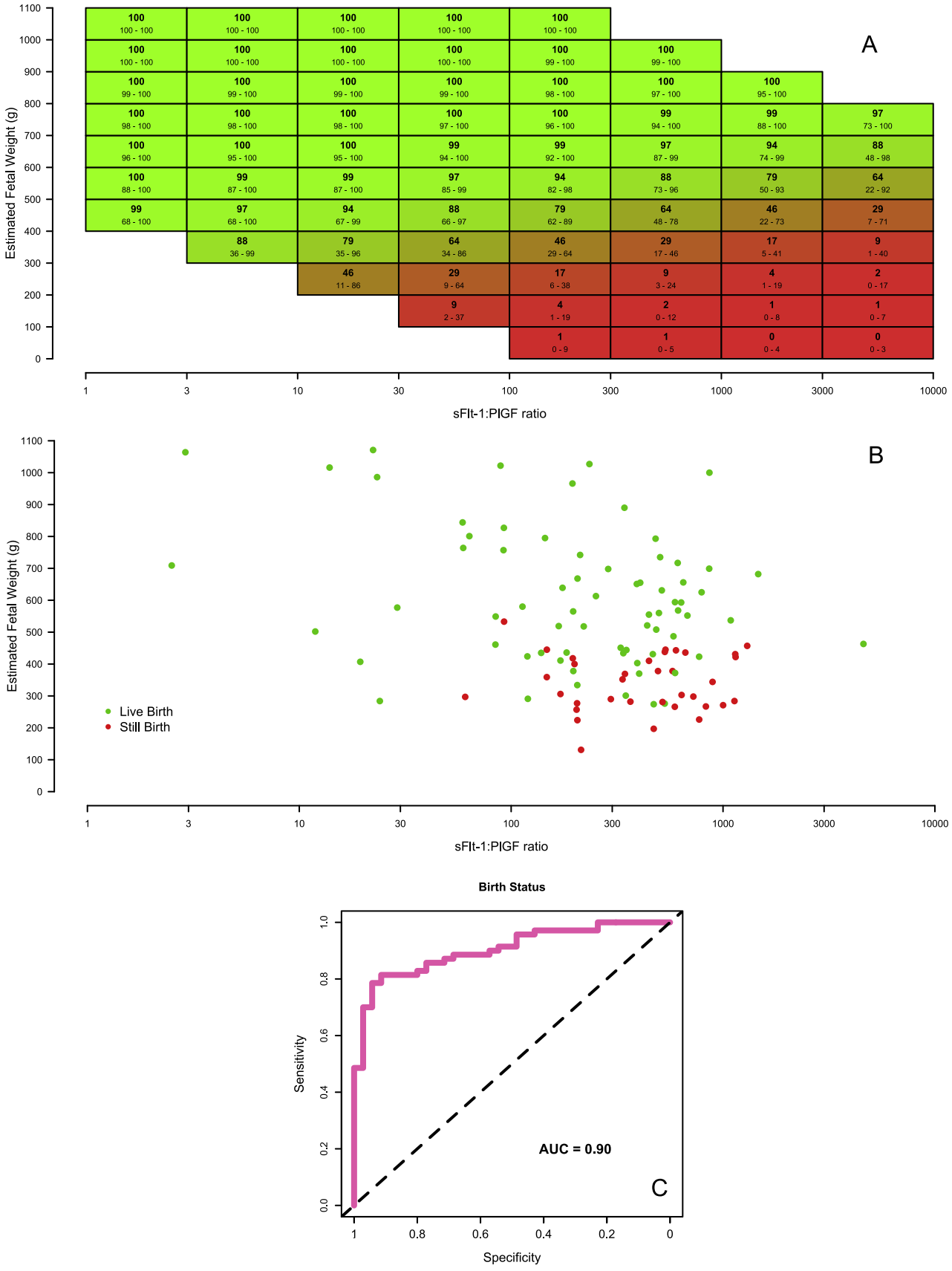

Fig. 2. Birth status by estimated fetal weight and sFlt-1:PlGF ratio at diagnosis. Data presented in graphical form denoting probability (95% CI) of a live birth (green denoted higher probability) (A), scatter graph of raw data (B) and ROC curve (C).

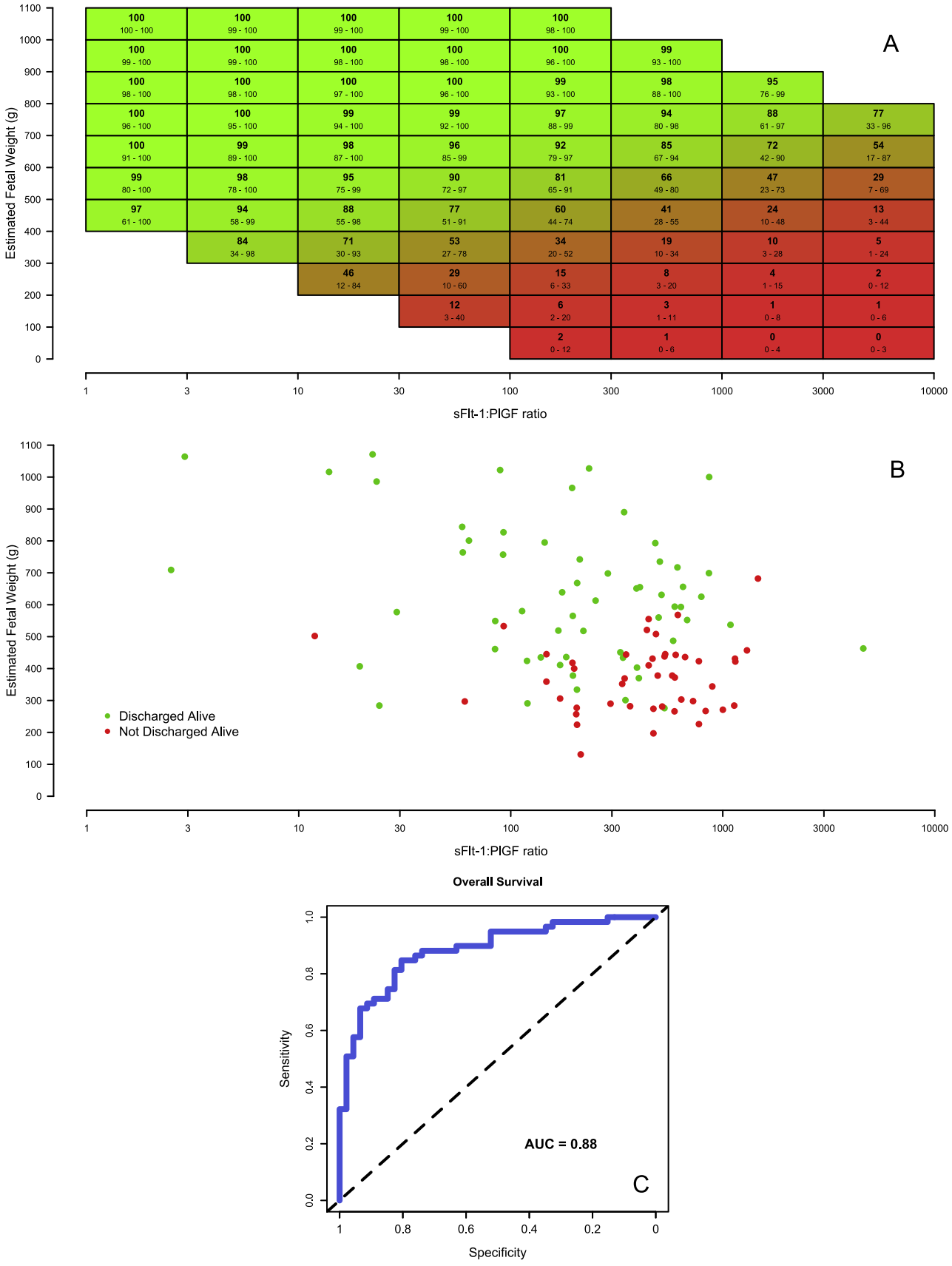

Fig. 3. Overall survival (discharged alive – green and perinatal mortality - red) by estimated fetal weight and sFlt-1:PlGF ratio at diagnosis. Data presented in graphical form denoting probability (95% CI) of a live discharge (green denoted higher probability) (A), scatter graph of raw data (B) and ROC curve (C).

severe early-onset FGR but does present a more detailed assessment of risk than has previously been performed. As such it has significant value in being able to guide clinicians and parents' decision making about the management of these high risk pregnancies. The prospective collection of rare and highly phenotyped cases managed within a single healthcare structure is unique.

This study has some limitations. Firstly some covariates (DV A-wave, UA PI) were not considered for inclusion in the modelling as the distribution of patients in these groups would not allow for reliable model estimates. Secondly, we did not mandate a clinical management pathway for FGR, which may mean that women were managed differently between units, but it may also provide 'real-life' evidence of fetal outcome in standard care. However, we would anticipate that the impact of this is minimal due to the large number of units all of whom would be guided by the national guidance within the UK [35].

Conclusion

This is the first time sFlt-1:PlGF ratio has been performed prospectively on a cohort of pregnancies complicated by severe early-onset FGR and correlated with a risk of adverse outcome in a prediction model. Use of this model may aid clinicians in determining the timing of fetal assessment and monitoring, timing of delivery, place of birth and may provide useful guidance for appropriate use of neonatal services. However, the main benefit of this model is likely to be in guiding the counselling of parents and families in the likelihood of a good outcome for their child.

Further large cohort studies will be required to validate this prediction model in severe early-onset FGR and as a predictive test for adverse pregnancy outcome including both short and long term physical and neurodevelopmental outcomes for the child.

STRIDER study group

Umber Agarwal, Elaine Willis and Silvia Mammarella, Liverpool Women's Hospital; Geraldine Masson, University Hospital of North Midlands, Stoke-on-Trent; Joe Aquilina, Elena Greco and Sally Higgins, The Royal London Hospital; Dimuthu Vinayagam and Louise Shaw, St. George's Hospital, London; Louise Stephens, St. Mary's Hospital, Manchester; David Howe and Abby Rand, Princess Anne Hospital, Southampton; Shalini Patni, Birmingham Heartlands Hospital; Tommy Mousa and Asma Rabab, Leicester Royal Infirmary; Helen Russell, NHS Fife Victoria Hospital, Scotland; Therese Hannon and Andrea Fenn, Royal Victoria Infirmary, Newcastle; Mark Kilby and Tara Selman, Birmingham Women's Hospital; Anna David and Rebecca Spencer, University College Hospital London; Kelly Cohen and Andrew Breeze, Leeds General Infirmary; Alastair McKelvey, Norfolk and Norwich University Hospitals, Lawrence Impey and Christos Loannou, John Radcliffe Hospital, Oxford; Sarah Stock, The Royal Infirmary of Edinburgh, Scotland; Liona Poon, Kings College London, Dharmintra Pasupathy and Louise Webster, St. Thomas' Hospital, London; and George Bugg, Nottingham City Hospital and Queen's Medical Centre.

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Ethical approval

Ethical approval was given by the North East Research Ethics Committee (14/NE/0011) in the United Kingdom. Each participating site provided a site specific approval and all participants signed a written informed consent.

The trial was sponsored by the University of Liverpool and Liverpool Women's Hospital. An Independent Data Monitoring Committee (IDMC) was established to review the safety and efficacy data. The protocol was registered on 31st July 2014, 4 months before the first patient was recruited (SRCTN39133303).

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Contribution to authorship

AS, RJ and ZA wrote the manuscript with which was subsequently critically reviewed and revised by all authors (CC, JH, MT, LK, PB, EJ, AK, PvD, AP). AS, CC and JH collated the data and RJ performed analysis and produced the data tables.

Declaration of Competing Interest

This study was not supported by Roche although AS has subsequently received Honoria for speaking at a Roche sponsored symposium. No other authors report any conflicts of interest

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Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:https://doi.org/10.1016/j.ejogrb.2019.08.007.

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