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Development of pre-eclampsia within 4 weeks of sFlt-1/PlGF ratio > 38: comparison of performance at 31–34 *vs* 35–37 weeks' gestation

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ABSTRACT

Objective *To compare the performance of screening by soluble fms-like tyrosine kinase 1 (sFlt-1) to placental growth factor (PlGF) ratio > 38 for the prediction of delivery with pre-eclampsia (PE) at < 1 week and < 4 weeks from assessment when the test is carried out at 31–34* vs *35–37 weeks' gestation.*

Methods *This was a prospective observational study in women attending a third-trimester ultrasound scan as part of routine pregnancy care; the visit was at 30–34 weeks' gestation in the first phase of the study and at 35–37 weeks in the second phase. Serum sFlt-1 and PlGF were measured and their ratio calculated. We estimated the detection rate (DR) and false-positive rate (FPR) of sFlt-1/PlGF ratio > 38 for predicting delivery with PE at < 1 week and < 4 weeks after assessment and compared the performance of screening when the test was carried out at 31 + 0 to 33 + 6* vs *35 + 0 to 36 + 6 weeks' gestation.*

Results *The study population included 8063 singleton pregnancies that were examined at 31–34 weeks and 3703 at 35–37 weeks. Delivery with PE occurred at < 1, < 4 and ≥ 4 weeks from assessment in five (0.1%), 29 (0.4%) and 202 (2.5%) women assessed at 31–34 weeks, respectively, and in seven (0.2%), 39 (1.1%) and 21 (0.6%) of those assessed at 35–37 weeks. In women without PE, the median sFlt-1/PlGF ratio increased with gestational age at screening and a ratio of 38 was just below the 99th percentile at 32 weeks' gestation and just below the 90th percentile at 36 weeks. In the two gestational windows, the DR of PE delivering < 4 weeks from assessment was similar (75.9% (95% CI, 56.5–89.7%)* vs *79.5% (95% CI, 63.5–90.7%)), but the FPR was substantially lower at 31–34 weeks than at 35–37 weeks (1.7% (95% CI, 1.4–2.0%)* vs *9.6% (95% CI, 8.7–10.6%)). The number of cases with PE delivering < 1 week from assessment was small, but similarly, in the two gestational windows, the DR was comparable (80.0% (95% CI, 28.4–99.5%)* vs *85.7% (95% CI, 42.1–99.6%)), and the FPR was substantially lower at 31–34 weeks than at 35–37 weeks (1.9% (95% CI, 1.6–2.2%)* vs *10.2% (95% CI, 9.3–11.3%)).*

Conclusion *The performance of sFlt-1/PlGF ratio > 38 in the prediction of delivery with PE at < 1 and < 4 weeks from assessment is substantially different when the assessment is at 31–34 weeks' gestation compared to at 35–37 weeks. Copyright © 2016 ISUOG. Published by John Wiley & Sons Ltd.*

INTRODUCTION

In women with pre-eclampsia (PE), the maternal serum concentration of the angiogenic placental growth factor (PlGF) is decreased and the level of antiangiogenic soluble fms-like tyrosine kinase-1 (sFlt-1) is increased[1,2]. There is also evidence that the altered levels of PlGF and sFlt-1 precede the clinical onset of the disease and a ratio of sFlt-1 to PlGF can be used in the assessment of women presenting to specialist clinics with signs or symptoms of hypertensive disorders to help distinguish between those who will develop PE in the subsequent 1–4 weeks from those who will not[3–8].

A prospective study in 1050 women with a singleton pregnancy presenting with signs or symptoms of hypertensive disorders at 24–37 (median, 32) weeks' gestation, reported that a sFlt-1/PlGF ratio ≤ 38 was best for predicting absence of PE at < 1 week from assessment and a ratio > 38 was best for predicting development of PE at < 4 weeks from assessment[8]. However, at 30–40 weeks in normal pregnancy, serum

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sFlt-1 increases and PlGF decreases with gestational age[2,9,10]; consequently, the sFlt-1/PlGF ratio would normally increase with gestational age and screening with a single cut-off point of 38 would result in an inevitable increase in the false-positive rate (FPR) with increasing gestational age.

The objective of this screening study in pregnant women undergoing routine clinical care at 31–37 weeks' gestation was to compare the performance of screening with sFlt-1/PlGF ratio > 38 in the prediction of delivery with PE at < 1, < 4 and ≥ 4 weeks from assessment when the test is carried out at 31–34 *vs* 35–37 weeks' gestation.

METHODS

The data for this study were derived from prospective screening for adverse obstetric outcomes in women attending a third-trimester routine hospital visit at King's College Hospital, London, or Medway Maritime Hospital, Gillingham, UK. In the first phase of the study, the visit was at 30–34 weeks' gestation and in the second phase the visit was at 35–37 weeks. The visits included recording of maternal demographic characteristics and medical history, ultrasound examination of fetal anatomy and growth, and measurement of serum PlGF and sFlt-1 in pg/mL using an automated biochemical analyzer (Cobas e411 system, Roche Diagnostics, Penzberg, Germany) within 10 min of blood sampling, with results being available 30 min later. Gestational age was determined by measurement of fetal crown–rump length at 11–13 weeks or fetal head circumference at 19–24 weeks[11,12].

Written informed consent was obtained from the women agreeing to participate in the study, which was approved by the NHS Research Ethics Committee. The inclusion criteria for this study were singleton pregnancy delivering a non-malformed live birth or stillbirth at ≥ 30 weeks' gestation in which sFlt-1 and PlGF were measured either at 31 + 0 to 33 + 6 or at 35 + 0 to 36 + 6 weeks' gestation. We excluded pregnancies with aneuploidy or major fetal abnormality. The study population was included in two previous reports[13,14].

Data on pregnancy outcome were collected from the hospital maternity records or the general medical practitioners of the women. The obstetric records of all women with pre-existing or pregnancy-associated hypertension were examined to determine if the condition was PE, as defined by the International Society for the Study of Hypertension in Pregnancy[15].

Statistical analysis

Percentiles were estimated using quantile regression[16]. We estimated and compared the detection rate (DR) and FPR, with their 95% CIs, of sFlt-1/PlGF ratio > 38 in the prediction of PE leading to delivery at < 1, < 4 and ≥ 4 weeks after assessment at 31 + 0 to 33 + 6 *vs* 35 + 0 to 36 + 6 weeks' gestation. The statistical software package R was used for data analyses[17] with the quantreg package[18] for quantile regression.

Table 1 Maternal and pregnancy characteristics of study population of women with singleton pregnancy assessed at 31–34 and 35–37 weeks' gestation for prediction of pre-eclampsia (PE)

Characteristic	*31–34 weeks (n = 8063)*	*35–37 weeks (n = 3703)*
PE delivering:		
< 1 week after assessment	5 (0.1)	7 (0.2)
< 4 weeks after assessment	29 (0.4)	39 (1.1)
≥ 4 weeks after assessment	202 (2.5)	21 (0.6)
Age (years)	31.0 (26.7–34.7)	31.7 (26.9–35.3)
Weight (kg)	77.0 (68.8–88.0)	79 (70.8–89.4)
Height (cm)	165 (160–169)	164 (160–169)
Racial origin		
Caucasian	6020 (74.7)	2748 (74.2)
Afro-Caribbean	1414 (17.5)	629 (17.0)
South Asian	305 (3.8)	135 (3.6)
East Asian	149 (1.8)	76 (2.1)
Mixed	175 (2.2)	115 (3.1)
Mode of conception		
Spontaneous	7792 (96.6)	3593 (97.0)
Assisted	271 (3.4)	110 (3.0)
Cigarette smoker	803 (10.0)	350 (9.5)
Chronic hypertension	119 (1.5)	45 (1.2)
APS/SLE	14 (0.2)	10 (0.3)
Diabetes mellitus		
Type 1	26 (0.3)	14 (0.4)
Type 2	53 (0.7)	17 (0.5)
Parity		
Nulliparous	3973 (49.3)	1735 (46.9)
Parous no previous PE	3788 (47.0)	1877 (50.7)
Parous previous PE	302 (3.7)	91 (2.5)
Family history of PE	243 (3.0)	133 (3.6)
Interpregnancy interval (years)*	3.2 (2.1–5.2)	3.1 (2.1–5.1)

Data are given as *n* (%) or median (interquartile range). *Interpregnancy interval reported for parous women. APS, antiphospholipid syndrome; SLE, systemic lupus erythematosus.

RESULTS

The study population included 8063 singleton pregnancies that were examined at 31–34 weeks' gestation and 3703 examined at 35–37 weeks. Delivery with PE at < 1, < 4 and ≥ 4 weeks after assessment occurred in five (0.1%), 29 (0.4%) and 202 (2.5%) women assessed at 31–34 weeks and in seven (0.2%), 39 (1.1%) and 21 (0.6%) of those assessed at 35–37 weeks. Maternal and pregnancy characteristics of the study population are summarized in Table 1.

The relationship between gestational age at assessment and sFlt-1/PlGF ratio is shown in Figure 1. In women without PE, the median sFlt-1/PlGF ratio increased significantly with gestational age at screening ($P < 0.0001$). A sFlt-1/PlGF ratio of 38 was just below the 99th percentile at 32 weeks' gestation and just below the 90th percentile at 36 weeks.

The DR and FPR of sFlt-1/PlGF ratio > 38 in the prediction of PE delivering at < 1, < 4 and ≥ 4 weeks after assessment at 31–34 weeks' gestation are compared to those after assessment at 35–37 weeks in Table 2. In the two gestational windows, the DR of PE delivering at < 4 weeks from assessment was similar (75.9% *vs* 79.5%),

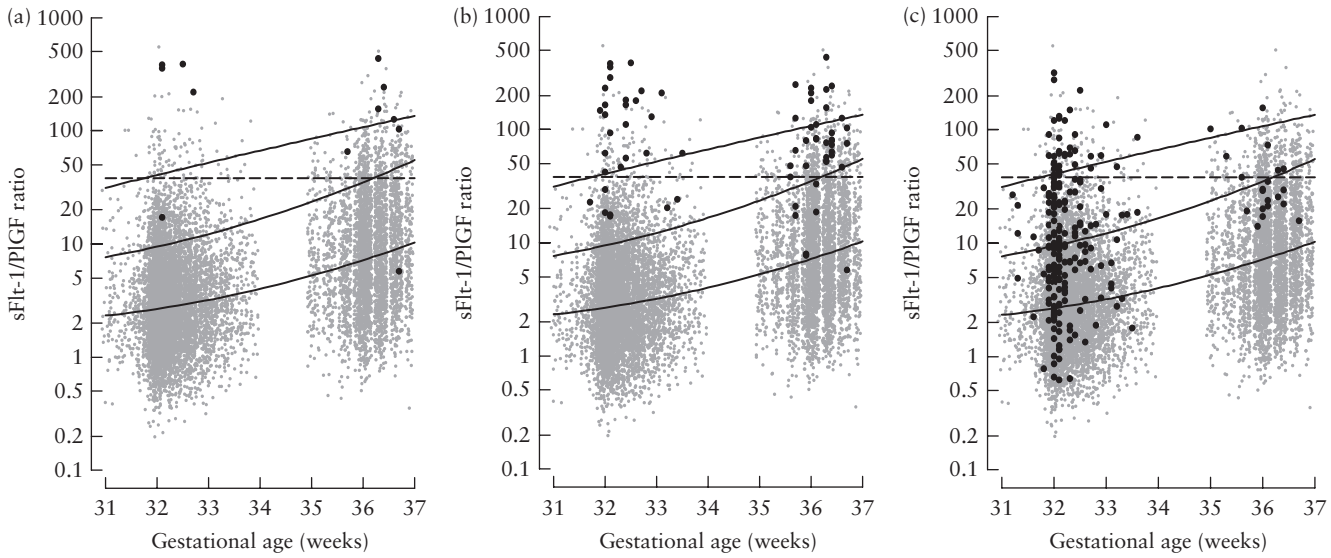

Figure 1 Serum soluble fms-like tyrosine kinase-1 to placental growth factor (sFlt-1/PlGF) ratio in singleton pregnancies with pre-eclampsia (PE) (•) that delivered < 1 week (a), < 4 weeks (b) or ≥ 4 weeks (c) after assessment at 31–34 or 35–37 weeks' gestation and in pregnancies that did not develop PE (•). Horizontal line represents sFlt-1/PlGF ratio cut-off point of 38 and curved lines represent 50th, 90th and 99th percentiles of unaffected pregnancies with gestational age.

but the FPR was substantially lower at 31–34 weeks than at 35–37 weeks (1.7% *vs* 9.6%). The number of cases with PE delivering at < 1 week from assessment was small, but similarly, in the two gestational windows, the DR was comparable (80.0% *vs* 85.7%), and the FPR was substantially lower at 31–34 weeks than at 35–37 weeks (1.9% *vs* 10.2%). In the prediction of PE delivering ≥ 4 weeks after assessment, the DR and FPR at 31–34 weeks (17.8% and 1.3%) were lower than at 35–37 weeks (38.1% and 9.5%).

DISCUSSION

Principal findings

The findings of this study demonstrate that, in normal pregnancy, there is a gestational age-related increase in the sFlt-1/PlGF ratio at 31–37 weeks' gestation. Consequently, in screening for PE with a fixed cut-off point for sFlt-1/PlGF ratio, there was an inevitable increase in the FPR with increasing gestational age; there was a five-fold increase in FPR with screening at 35–37 weeks compared to screening at 31–34 weeks. In contrast, in pregnancies delivering with PE at < 1 and < 4 weeks from assessment, there was a similar increase in sFlt-1/PlGF ratio from the fixed cut-off point and therefore the DR for screening at 31–34 weeks was similar to that for screening at 35–37 weeks. The performance of screening for PE delivering at ≥ 4 weeks from assessment was poor at both 31–34 weeks and at 35–37 weeks.

Strengths and limitations

The strengths of this screening study for PE in the third trimester of pregnancy are first, examination of a large population of women attending for routine care, second, measurement of serum sFlt-1 and PlGF by automated machines that provide reproducible results within 40 min of sampling so that complete assessment and counseling can potentially be undertaken in the same hospital visit, and third, use of sFlt-1/PlGF ratio cut-off that was proposed and validated previously for prediction of PE at < 1 and < 4 weeks after assessment at 24–37 weeks' gestation[8].

A potential limitation of the study relates to the objective of comparing the performance of sFlt-1/PlGF ratio > 38 in an unselected population, in which the prevalence of PE was about 3%, to that in women presenting to specialist clinics with signs or symptoms of hypertensive disorders in which the prevalence of PE was 18%[8]. However, the DR of the test for PE delivering at < 1 and < 4 weeks from assessment was similar because the DR is not affected by the prevalence of PE.

Comparison with previous studies

The finding that in normal pregnancy during the third trimester the sFlt-1/PlGF ratio increases with gestational age is comparable with previous reports[2,9,10].

A previous study in 1050 singleton pregnancies presenting with signs or symptoms of hypertensive disorders at 24–37 weeks' gestation selected the sFlt-1/PlGF ratio with the single cut-off point of 38 to predict PE delivering at < 1 and < 4 weeks from assessment because of its simplicity and because its performance was apparently similar to that of a model with two cut-off points, one for the earlier gestational phase of 24 to 34 weeks and one for the later gestational phase of ≥ 34 weeks, and a model with a different cut-off point for each gestational week[8]. However, the study did not provide details on how the performance of the three models was evaluated

Table 2 Performance of serum soluble fms-like tyrosine kinase-1 to placental growth factor (sFlt-1/PlGF) ratio > 38 in prediction of delivery with pre-eclampsia (PE) < 1, < 4 and ≥ 4 weeks after assessment at 31–34 or 35–37 weeks' gestation

	GA at assessment (weeks)	*PE with delivery:*		
		< 1 week	*< 4 weeks*	*≥ 4 weeks*
Detection rate	31–34	4/5 (80.0 (28.4–99.5))	22/29 (75.9 (56.5–89.7))	36/202 (17.8 (12.8–23.8))
	35–37	6/7 (85.7 (42.1–99.6))	31/39 (79.5 (63.5–90.7))	8/21 (38.1 (18.1–61.6))
False-positive rate	31–34	152/8058 (1.9 (1.6–2.2))	134/8034 (1.7 (1.4–2.0))	98/7832 (1.3 (1.0–1.5))
	35–37	378/3696 (10.2 (9.3–11.3))	353/3664 (9.6 (8.7–10.6))	345/3643 (9.5 (8.5–10.5))

Data are given as *n/N* (% (95% CI)). GA, gestational age.

and found to be similar. Our study showed that, although the DR of PE delivering at < 1 and < 4 weeks from assessment with screening by sFlt-1/PlGF ratio > 38 was similar with screening at 31–34 and at 35–37 weeks, the FPR at 35–37 weeks was five times higher than at 31–34 weeks.

Implications for clinical practice

In third-trimester screening for PE, serum sFlt-1 and PlGF are powerful biomarkers for PE delivering at < 4 weeks from assessment and their individual performance of screening is superior to that of uterine artery pulsatility index and mean arterial pressure, which are two other useful biomarkers; however, the performance of a model that combines maternal characteristics and medical history with all four biomarkers is superior to that which combines only sFlt-1 and PlGF[13,14,19].

The sFlt-1/PlGF ratio as a method of screening for PE in both the general population and in high-risk pregnancies is attractive because of its simplicity and the performance of screening for PE delivering at < 1 and < 4 weeks is similar to that of our preferred method of utilizing Bayes' theorem to combine the prior risk from maternal characteristics and medical history with multiples of the median (MoM) values of sFlt-1 and PlGF to derive the patient-specific posterior risks. However, if sFlt-1/PlGF ratio, rather than the method that uses Bayes' theorem, was to be applied, a high performance in screening necessitates the use of a variable rather than a fixed ratio. A precondition for effective use of a fixed cut-off is that the distribution of values in both the PE and unaffected cases does not change with gestation; as demonstrated in this study this is not the case for unaffected pregnancies. Consequently, use of sFlt-1/PlGF ratio > 38 in identifying a high-risk group in need of intensive monitoring in the subsequent 4 weeks would lead to five times more pregnancies being classified falsely as high risk when screening is carried out at 35–37 weeks than at 31–34 weeks.

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