Journal of Biotechnology 300 (2019) 11–19

Contents lists available at ScienceDirect

Journal of Biotechnology

journal homepage: www.elsevier.com/locate/jbiotec

Screening for preeclampsia in the first trimester of pregnancy in routine clinical practice in Hungary

Laszlo Orosz[a], Gergo Orosz[a], Lajos Veress[b], Diana Dosa[c], Laszlo Orosz Sr[d], Ibolya Arany[e], Antal Fabian[d], Laszlo Medve[d], Karoly Pap[d], Zsolt Karanyi[f], Zoltan Toth[a], Robert Poka[a], Nandor Gabor Than[g,h,i,**], Olga Torok[a,*]

[a] Department of Obstetrics and Gynaecology, Faculty of Medicine, University of Debrecen Medical and Health Science Centre, Nagyerdei korut 98, 4032, Debrecen, Hungary
[b] Department of Pharmacology and Pharmacotherapy, Faculty of Medicine, University of Debrecen Medical and Health Science Centre, Nagyerdei korut 98, 4032, Debrecen, Hungary
[c] Department of Family and Occupational Medicine, Faculty of Public Health and Faculty of Medicine, University of Debrecen Medical and Health Science Centre, Moricz Zsigmond krt. 22, 4032, Debrecen, Hungary
[d] Departement of Obststrics and Gynaecology, Andras Josa County and Teaching Hospital, Szent Istvan ut. 68, 4400, Nyiregyhaza, Hungary
[e] Departement of Neonatology, Andras Josa County and Teaching Hospital, Szent Istvan ut. 68, 4400, Nyiregyhaza, Hungary
[f] Department of Internal Medicine, Faculty of Medicine, University of Debrecen Medical and Health Science Centre, Nagyerdei korut 98, 4032, Debrecen, Hungary
[g] Systems Biology of Reproduction Lendulet Research Group, Institute of Enzymology, Research Centre for Natural Sciences, Hungarian Academy of Sciences, Magyar Tudosok krt. 2, 1117, Budapest, Hungary
[h] Maternity Private Clinic of Obstetrics and Gynecology, Kiralyhago ter 8, 1126, Budapest, Hungary
[i] First Department of Pathology and Experimental Cancer Research, Semmelweis University, Ulloi ut 26, 1085, Budapest, Hungary

ARTICLE INFO

Keywords:
First trimester
Preeclampsia
Screening
Uterine artery Doppler
Blood pressure
Placental growth factor

ABSTRACT

We aimed to evaluate the contribution of different factors in the Fetal Medicine Foundation algorithms for preeclampsia (PE) risk calculation during first-trimester screening in Hungary. We selected subjects for the nested case-control study from a prospective cohort of 2545 low-risk pregnancies. Eighty-two patients with PE and 82 gestational age-matched controls were included. Individual PE risk was calculated using two risk-assessing softwares. Using Astraia 2.3.1, considering maternal characteristics and biophysical parameters only, detection rates (DR) were 63.6% for early-PE and 67.6% for late-PE. When we added placenta associated plasma protein A (PAPP-A) to the risk calculation, DRs decreased to 54.5% and 64.8% respectively. Using Astraia 2.8.2 with maternal characteristics and biophysical parameters resulted in the DRs of 63.6% (early-PE) and 56.3% (late-PE). If we added PAPP-A to the risk calculation, DRs improved to 72.7% and 54.9%. The addition of placental growth factor (PlGF) did not increase detection rates in either calculation. In conclusion, using maternal characteristics, biophysical parameters, and PAPP-A, an acceptable screening efficacy could be achieved for early-PE during first-trimester screening. Since PlGF did not improve efficacy in our study, we suggest setting new standard curves for PlGF in Eastern European pregnant women, and the evaluation of novel biochemical markers.

Abbreviations: APS, antiphospholipid syndrome; A1, Astraia software 2.3.2; A2, Astraia software 2.8.1; BhCG, beta human choriogonadotropin; BPD, biparietal diameter; BP, blood Pressure; ctrl, control; CRL, crown-rump length; DR, detection rate; DM, diabetes mellitus; DV-PI, ductus venosus pulsability index; PET, family history of preeclampsia; FPR, false-positive rate; FHR, fetal heart rate; FMF, Fetal Medicine Foundation; IVF, in vitro fertilization; a priori risk, maternal background risk; MAP, mean arterial pressure; MoM, multiple of expected median; NT, nuchal translucency; PE, preeclampsia; PAPP-A, placenta associated plasma protein A; PlGF, placental growth factor; SGA, small for gestational age; SLE, systemic lupus erythematosus; UtA-PI, uterine artery pulsatility index; vs, versus; wks, weeks; yrs, years

* Corresponding author.
** Corresponding author at: Systems Biology of Reproduction Lendulet Research Group, Institute of Enzymology, Research Centre for Natural Sciences, Hungarian Academy of Sciences, Magyar Tudosok krt. 2, 1117, Budapest, Hungary.
E-mail addresses: oroszlaszlo@med.unideb.hu (L. Orosz Sr), than.gabor@ttk.mta.hu (N.G. Than), to@med.unideb.hu (O. Torok).

https://doi.org/10.1016/j.jbiotec.2019.04.017
Received 31 January 2019; Received in revised form 18 April 2019; Accepted 20 April 2019
Available online 02 May 2019

1. Introduction

Preeclampsia (PE) prevalence is approximately 2–4% of all pregnancies (Duley, 2009). In developed countries PE is one of the leading causes of maternal morbidity (Mayrink et al., 2018) and responsible for approximately 14% of all pregnancy-related deaths (Chaiworapongsa et al., 2014a). From a fetal point of view, PE is associated with an increased risk of perinatal morbidity and mortality, responsible for approximately 10% of stillbirths (Gardosi et al., 2005) and 12–16% of preterm births (Dhariwal and Lynde, 2017). In the last decade, extensive efforts have been made to develop an efficient screening method for preeclampsia with the aim to reduce its prevalence through pharmacologic intervention in the high-risk group (Chaiworapongsa et al., 2014b; Rolnik et al., 2017a, 2017b). The traditional screening based on risk factors from maternal medical history can identify only 35% of all preeclampsia cases and approximately 40% of early-PE cases at a false-positive rate (FPR) of 10% (Wright et al., 2015). Over the past years, several studies have proved that a combination of previous maternal history with specific biophysical and biochemical markers can predict PE in the first trimester (Baschat et al., 2014; Wright et al., 2019a), and this screening method is superior to that of using maternal characteristics only (O'Gorman et al., 2017). Previous studies have also shown that this is the only period of the pregnancy when preventive strategies with aspirin proved to be effective (Rolnik et al., 2017b; Wright and Nicolaides, 2019b). Screening for early-PE has a higher sensitivity than for late-PE. Detection rates (DR) for early-PE range from 41% (Poon et al., 2009a) to 96% (Foidart et al., 2010), depending on the markers and algorithms used in the risk calculation. DRs for late-PE range between 31–45% (Poon et al., 2009b; Erez et al., 2017). Studies on PE screening have been performed mostly on Anglo-Saxon populations and have almost similar research settings, and thus, have comparable results (Poon et al., 2009c, 2010a, 2010b). Studies carried out on South European or Middle European population with smaller sample sizes could not identify any subgroups as early- or late-PE (Pilalis et al., 2007). More studies have been suggested in the literature in populations that are different from those of the original investigations (Scazzocchio et al., 2013; Agarwal et al., 2017) because the contribution of different races and their lifestyles have a significant impact on the maternal background risk (a priori risk). These differences might also influence the interpretation of the measurement of the different biophysical and biochemical markers. For example, in Eastern European countries, obesity is less common but cardiovascular risk factors are present almost with the same if not higher (Timmis et al., 2017) frequency as in the United Kingdom (Tunstall-Pedoe et al., 1999). These findings and factors necessitate further studies to examine the efficacy of first-trimester screening for PE in routine clinical practice. In this study, our aim was to evaluate the effectiveness of first trimester screening for PE under routine clinical practice in an Eastern European unselected population using Fetal Medicine Foundation (FMF) softwares (Lobo et al., 2019) and to examine the contribution of different factors in the algorithms offered by FMF for risk calculation.

2. Materials and methods

2.1. Recruitment of patients

We performed a prospective cohort study at the Department of Obstetrics and Gynaecology at the University of Debrecen Medical and Health Science Centre, Debrecen, Hungary and the Department of Obstetrics and Gynaecology at the Andras Josa County and Teaching Hospital, Nyiregyhaza, Hungary. We recruited participants at the time of the routine first-trimester screening for fetal malformations and chromosomal abnormalities. The local ethics committee approved the study protocol (identification number: DEOEC RKEB/IKEB 3092-2010), and each patient gave written informed consent to participation. Gestational age was determined by the crown-rump length (CRL) at first-trimester scan (Robinson et al., 1979). Pregnant women with gestational age between 11^{+0} and 13^{+6} weeks were included.

2.2. Recording maternal characteristics — a priori risk

Before the first-trimester ultrasound scan, a patient questionnaire on maternal characteristics and previous medical history recommended for the use of FMF algorithm for preeclampsia risk calculation had to be filled in (Table 1). The questionnaire was then reviewed by a physician together with the patient.

2.3. Biophysical and biochemical measurements

Blood pressure (BP) was measured using FMF guidelines (Gallo et al., 2014) with a calibrated device (M2 Intellisense; Omron Corp, Kyoto, Japan). The first-trimester ultrasound screening and uterine artery pulsatility index (UtA-PI) evaluation was performed transabdominally according to FMF guidelines (Khalil et al., 2014) by physicians with FMF license. We recorded CRL, nuchal translucency (NT), biparietal diameter (BPD), fetal heart rate (FHR), ductus venosus pulsatility index (DV-PI), tricuspid valve assessment, UtA-PI. Blood (serum and plasma) and urine samples were collected before the scan and stored at −80 °C degrees for further studies. For the current study, maternal serum beta human choriogonadotropin (BhCG), placenta associated plasma protein A (PAPP-A) and placental growth factor (PlGF) serum levels were measured using a BRAHMS Kryptor analyzer (ThermoFisher). Since the normal serum levels of BhCG, PAPP-A and PLGF are influenced by several factors such as parity, gestational age, maternal BMI, race, method of conception and smoking status, these biochemical results were converted to multiples of the expected normal median (MoM) by the FMF algorithm. Using MoMs, the standardized maternal serum levels of these biomarkers could be compared later.

Table 1
Patient questionnaire of previous medical history. Years (yrs), diabetes mellitus (DM), systemic lupus erythematosus (SLE), antiphospholipid syndrome (APS), in vitro fertilization (IVF), small for gestational age (SGA).

Questions (maternal)	Possible answers				
age (yrs), weight (kg), height (cm)	specific number				
racial origin	Caucasian	Afro-Caribbean	South Asian	East Asian	mixed
method of conception	spontaneous	ovulation induction	IVF		
cigarette smoking	yes	no			
chronic hypertension	yes	no			
maternal type 1 or 2 DM	yes	no			
maternal SLE	yes	no			
maternal APS	yes	no			
family history of PE in the mother of the patient	yes	no			
parity	nulliparous	parous			
		previous pregnancy with PE	yes	no	
		previous pregnancy with SGA babies	yes	no	

Table 2
Characteristics of the study population of pregnant women screened between 11^{+0} and 13^{+6} weeks for PE. Years (yrs), weeks (wks), preeclampsia (PE).

Characteristic	Unaffected (n = 82)	PE X < 34wks (n = 11)	PE 34 < X < 37wks (n = 11)	PE X > 37wks (n = 60)
Maternal age (yrs)	28.9 (17.7–39.5)	28.1 (15.8-37.9)	29.7 (23.4–38.5)	28.5 (18.4-38.6)
Maternal weight (kg)	62.6 (45–98)	74.8 (46–109)	77.6 (54–93)	74.4 (43-124)
Maternal height (cm)	165.0 (154–180)	162.3 (156–176)	169.1 (156–182)	165.9 (150-184)
Body Mass Index (kg/m2)	22.9 (17–32)	28.2 (18–44.2)	27.9 (18–35.2)	26.9 (17–46.7)
Gestational age (wks)	12.7 (11.3–13.9)	12.6 (11.7–13.4)	12.4 (11.6–13.3)	12.8 (11.3–13.9)
Racial origin				
Caucasian	82 (100%)	11 (100%)	11 (100%)	60 (100%)
Medical history				
Chronic hypertension	0 (0%)	1 (9%)	5 (45%)	15 (25%)
Diabetes mellitus	0 (0%)	1 (9%)	2 (18%)	0 (0%)
Mode of conception				
Spontaneous	82 (100%)	10 (91%)	11 (100%)	58 (97%)
In-vitro fertilization	0 (0%)	0 (0%)	0 (0%)	2 (3%)
Ovulation induction	0 (0%)	1 (9%)	0 (0%)	0 (0%)
Parity				
Nulliparous	37 (45%)	8 (73%)	8 (73%)	44 (73%)
Parous	45 (55%)	3 (27%)	3 (27%)	16 (27%)
No previous PE	45 (55%)	3 (27%)	1 (9%)	13 (22%)
Previous PE	0 (0%)	0 (0%)	2 (18%)	3 (5%)

2.4. Follow up of the study population

Between 2010 and 2012, we recruited a total of 2545 pregnant women with the aim of finding further useful biochemical markers for the screening of major adverse pregnancy complications as pre-eclampsia, macrosomia, fetal growth restriction and gestational diabetes. In the current nested case-control study, we focused on pre-eclampsia. The outcomes were collected from the electronical medical database of each center (MedSol-Debrecen, MedWorks-Nyiregyhaza). Those patients who did not deliver in either of these centers received phone calls and their medical files were sent to us by their local hospital. Out of the 2545 patients, in 294 cases the outcome could not be collected due to the loss of follow-up. From the remaining 2251 pregnancies, 2223 resulted in live birth. There were nine terminations for chromosomal abnormalities or multiplex malformations, 11 miscarriages, and eight intrauterine deaths. For this study, we used data only from singleton pregnancies, and in consequence 23 multiple pregnancies were excluded out of the 2223 pregnancies. In the remaining 2200 singleton cases, 82 ended up with PE (3.7%, 82/2200). Two medical doctors double checked all the outcome data of those patients diagnosed with PE. We defined patients with early-PE as those with PE requiring delivery before 34 weeks. Late-PE was defined as PE requiring delivery after 34 weeks (Bahado-Singh et al., 2015). The rate of early-PE was 0.5% (11/2200) and 3.2% (71/2200) of late-PE. For this study, 82 uncomplicated low-risk pregnancies were selected as controls, matched for gestational age and maternal age at sample collection. These were compared with the 82 PE cases to assess the role of the biochemical markers recommended by two different FMF algorithms in the screening efficacy for preeclampsia during routine clinical practice.

2.5. Calculating the risk of PE using different algorithms

We used two commercially available softwares for the risk calculation, Astraia 2.3.2 (A1, launched in 2010) and Astraia 2.8.1 (A2, launched in 2016, Astraia Software GmBH, Occamstr. 20, 80802, Munich, Germany). For the calculation of a priori risk for PE, A1 software required the ethnic origin, parity, maternal weight, maternal height, chronic hypertension, smoking status, method of conception, family history of preeclampsia (PET), maternal BP and UtA-PI. The A1 program gave two types of risks: 1) Risk for early-PE (before 34 weeks) and 2) Risk for late-PE (after 34 weeks). In the general population, the risk for early-PE is 0.5% and 2% for late-PE (Poon et al., 2009b), so we defined screen-positive patients as those with a risk of > 0.5% for early-PE and risk of > 2% for late-PE. For a priori risk calculation for PE, the new A2 software required the same data as A1 as well as four additional maternal risk factors: maternal diabetes mellitus (DM), systemic lupus erythematosus (SLE), antiphospholipid syndrome (APS), and previous SGA newborn. Modified mathematical algorithms were used by A2 software compared with A1 software to calculate the mean arterial pressure (MAP), MAP MoM, and UtA-PI MoM. The results for PE risk were divided into three subgroups by the A2 program: 1) Preeclampsia before 34 weeks (early-PE); 2) Preeclampsia between 34–37 weeks; and 3) Preeclampsia after 37 weeks. Groups 2 and 3 together formed the late-PE group. We calculated the risk for PE for the 82 PE patients, and the 82 controls by both A1 and A2 programs and compared the results. For both programs the risks were calculated in three ways: 1) Using maternal characteristics and biophysical measurements alone; 2) Using maternal characteristics, biophysical measurements, and PAPP-A; and 3) Using maternal characteristics, biophysical measurements, PAPP-A and PlGF (PlGF could be used by A2 software only).

3. Calculation

We used the R-project v.3.5., a software package (R Core Team, 2018) for statistical analysis. Descriptive statistics with median, quartiles, mean, standard deviation and case number were used to characterized variables. We performed logarithmic transformation of parameters in case of variables with non-normal distribution. We used unpaired student's *t*-test and one-way analysis of variance (ANOVA) for comparisons between values of groups. Tukey-HSD posthoc test was used to corroborate ANOVA results. Student's paired *t*-test was used to assess the difference between the versions of parameters (MAP, MAP MoM, UtA-PI). We examined explanatory variables using ROC curve analysis (pROC package) (Xavier R, 2011) and generated optimal thresholds and AUCs. During the calculations, we accepted $p <= 0.05$ probability levels as significant.

4. Results

4.1. Characteristics of the study population

Table 2 presents the maternal characteristics of the study population.

In the early-PE group the rate of chronic hypertension (1) and previous history of PE (0) was relatively low compared with the other groups of PE.

Table 3 shows the distribution of the markers used for the risk

Table 3
Distribution of the markers used in the risk calculation for PE and the differences between the calculation of MAP, MAP MoM and UtA-PI MoM by the A1 and A2 programs. We highlighted comparisons with significant differences. Blood pressure (BP), multiple of expected median (MoM), mean arterial pressure (MAP), uterine artery pulsatility index (UtA-PI), beta human choriogonadotropin (BhCG), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF), Astraia software 2.3.2 (A1), Astraia software 2.8.1 (A2).

	Total (n = 164)					Test A1 vs A2 p-value
	25th centile	median	75th centile	mean	sd	
Age (yrs)	25.7	28.6	32.4	28.8	4.7	
Weight (kg)	55.8	66.0	78.3	68.7	16.3	
Height (cm)	160.0	165.0	170.0	165.4	6.5	
BMI (kg/m^2)	21.0	23.5	28.2	25.1	5.5	
BP MoM	1.030	1.070	1.150	1.088	0.102	
A1 software MAP	87.10	91.80	102.13	94.55	10.50	**A1 vs A2 MAP**
A2 software MAP	85.98	91.05	100.25	92.89	10.22	**< 0.0001**
A1 software MAP MoM	1.045	1.096	1.189	1.119	0.110	**A1 vs A2 MAP MoM**
A2 software MAP MoM	1.030	1.070	1.150	1.088	0.102	**< 0.0001**
A1 software UtA-PI MoM	0.882	1.023	1.180	1.060	0.247	**A1 vs A2 UtA-PI MoM**
A2 software UtA-PI MoM	0.870	0.970	1.160	1.019	0.223	**< 0.0001**
BhCG MoM	0.668	0.995	1.459	1.195	0.774	
PAPP-A MoM	0.816	1.141	1.548	1.317	0.816	
PlGF MoM	0.979	1.239	1.678	1.387	0.651	

calculation in the whole population and demonstrates the differences between the mathematical algorithms used by A1 and A2 programs for the calculation of MAP, MAP MoM, and UtA-PI MoM.

4.2. Comparison of the characteristics within the groups

Tables 4a and 4b shows that when comparing the characteristics of the PE and the control (ctrl) group, there were differences for maternal weight, BMI, BP MoM, UtA-PI MoM, PlGF MoM, PAPP-A MoM. Although PlGF levels were lower in the PE group compared to controls in our study population, the median of the PlGF MoMs was shifted to the right in both groups by the FMF algorithm calculation (Fig. 1).

Using the receiver–operating characteristics curves (Fig. 2), only BP MoM, UtA-PI MoM, and BMI showed a significant contribution to the prediction of PE.

4.3. Characteristics of the PE subgroups and the control population

The risk for PE was divided into three subgroups according to the calculation of A2 software as 1) early-PE PE < 34 wks (PE < 34); 2) PE between 34–37 wks (PE 34–37); and 3) PE after 37 wks (PE > 37). The PlGF MoM, PAPP-A MoM, BP MoM, UtA-PI MoM, BhCG MoM, and maternal characteristics were compared within all three subgroups and with the control group (ctrl) (Tables 4a and 4b). There were differences for maternal weight between ctrl vs. PE 34–37 and ctrl vs. PE > 37 groups. Regarding BMI, there were differences between all three subgroups of PE when compared to controls. The BP MoMs showed similar differences as BMI, and also between PE > 37 vs. PE34–37 groups. There was UtA-PI difference between ctrl vs. PE < 34 and ctrl vs. PE > 37 groups. There was a difference regarding PAPP-A MoMs between groups ctrl vs. PE < 34, meanwhile PlGF MoM results showed a difference between ctrl vs. PE34–37 groups only.

4.4. Detection rates of A1 and A2 programs using different settings

Table 5 shows the screening efficacy of the two programs in different settings.

With A1 software, out of the 11 cases of early-PE, 7 (63.6%, 7/11) were screen positive (risk ›1:200) using the maternal a priori risk, UtA-PI and BP measurements without biochemistry and only 6 (54.5%, 6/11) when adding PAPP-A to the risk calculation. Out of the 71 patients with late-PE, the risk was high (risk ›1:50) in 48 cases (67.6%, 48/71) without PAPP-A and in 46 cases (64.8%, 46/71) with PAPP-A, respectively. The false positive rate among the 82 controls was 6%, (5/82) in both subgroups of A1 software.

With A2 software, out of the 11 early-PE cases, 7 (63.6%, 7/11) were screen positive without biochemical results. Eight cases (72.7%, 8/11) were identified when PAPP-A was added to the risk calculation. Adding PlGF as a further biochemical marker to the risk calculation resulted in a drop of the detection to 5 cases (45.5%, 5/11) in the early-PE group. In the group where PE developed between 34–37 weeks, 10 cases were screen positive (90.1%, (10/11) either with or without PAPP-A and 8 cases (72.7%, (8/11) when we added PlGF to the risk calculation. In the group where PE developed after 37 weeks, the

Table 4a
Pregnancy characteristics in PE compared with the control group. We highlighted the comparisons with significant differences. Years (yrs), mean arterial pressure (MAP), multiple of expected median (MoM), uterine artery pulsatility index (UtA-PI), beta human choriogonadotropin (BhCG), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF).

	PE (n = 82)					Control (ctrl, n = 82)					Test (PE vs ctrl)
	25th centile	Median	75th centile	Mean	sd	25th centile	Median	75th centile	Mean	sd	p-value
Age (yrs)	25.5	28.6	32.3	28.6	4.9	25.8	28.6	32.5	29.0	4.6	0.5231
Weight (kg)	62.0	72.5	88.0	74.9	16.9	54.0	60.0	69.0	62.6	13.0	**< 0.0001**
Height (cm)	160.0	167.0	170.0	165.9	7.1	160.0	165.0	168.8	165.0	5.9	0.4304
BMI (kg/m^2)	23.0	26.5	31.0	27.2	5.6	20.0	22.0	25.0	22.9	4.4	**< 0.0001**
MAP MoM	1.080	1.145	1.218	1.142	0.104	1.003	1.040	1.070	1.035	0.067	**< 0.0001**
UtA-PI MoM	0.920	1.045	1.230	1.085	0.242	0.840	0.930	1.070	0.951	0.181	**< 0.0001**
bHCG MoM	0.635	1.035	1.501	1.246	0.895	0.722	0.947	1.430	1.145	0.633	0.8339
PAPP-A MoM	0.685	1.065	1.453	1.207	0.828	0.947	1.176	1.784	1.426	0.794	**0.0068**
PlGF MoM	0.844	1.215	1.551	1.280	0.632	1.046	1.284	1.889	1.494	0.655	**0.0118**

Table 4b
Maternal, biophysical and biochemical characteristics in the three subgroups of PE compared with controls. We highlighted the comparisons with significant differences. Years (yrs), body mass index (BMI), blood pressure (BP), multiple of expected median (MoM), uterine artery pulsatility index (UtA-PI), beta human choriogonadotropin (BhCG), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF), preeclampsia (PE), control (ctrl).

	PE	n	25th centile	Median	75th centile	Mean	sd	Test	ANOVA
Age (yrs)	PE < 34	11	23.01	26.50	34.90	28.06	7.37	PE34-37 vs PE < 34	0.8620
	PE 34-37	11	24.58	29.08	33.63	29.66	5.41	PE > 37 vs PE < 34	0.9944
	PE > 37	60	26.54	28.69	31.62	28.45	4.29	Ctrl vs PE < 34	0.9309
	Ctrl	82	25.83	28.60	32.45	28.99	4.62	PE > 37 vs PE34-37	0.8690
								Ctrl vs PE34-37	0.9721
								Ctrl vs PE > 37	0.9132
Weight (kg)	PE < 34	11	57.50	72.00	90.00	74.82	21.85	PE34-37 vs PE < 34	0.8982
	PE 34-37	11	69.50	80.00	87.50	77.64	12.71	PE > 37 vs PE < 34	0.9995
	PE > 37	60	62.00	71.50	87.25	74.38	16.85	Ctrl vs PE < 34	0.0933
	Ctrl	82	54.00	60.00	69.00	62.62	12.98	PE > 37 vs PE34-37	0.8558
								Ctrl vs PE34-37	**0.0072**
								Ctrl vs PE > 37	**0.0000**
Height (cm)	PE < 34	11	157.50	160.00	167.50	162.27	6.50	PE34-37 vs PE < 34	0.0673
	PE 34-37	11	164.50	171.00	173.50	169.09	7.42	PE > 37 vs PE < 34	0.3119
	PE > 37	60	162.00	167.00	170.00	165.93	6.98	Ctrl vs PE < 34	0.5503
	Ctrl	82	160.00	165.00	168.75	165.01	5.90	PE > 37 vs PE34-37	0.4449
								Ctrl vs PE34-37	0.2044
								Ctrl vs PE > 37	0.8352
BMI (kg/m^2)	PE < 34	11	23.15	25.50	32.10	28.23	7.50	PE34-37 vs PE < 34	1.0000
	PE 34-37	11	25.50	27.80	30.95	27.87	4.98	PE > 37 vs PE < 34	0.9371
	PE > 37	60	22.95	26.15	30.30	26.92	5.41	Ctrl vs PE < 34	**0.0094**
	Ctrl	82	20.03	22.00	24.95	22.95	4.38	PE > 37 vs PE34-37	0.9268
								Ctrl vs PE34-37	**0.0084**
								Ctrl vs PE > 37	**0.0000**
BP MoM	PE < 34	11	1.0750	1.1400	1.1850	1.1527	0.0980	PE34-37 vs PE < 34	0.4761
	PE 34-37	11	1.1450	1.1900	1.2800	1.2055	0.0919	PE > 37 vs PE < 34	0.8131
	PE > 37	60	1.0775	1.1350	1.2000	1.1278	0.1036	Ctrl vs PE < 34	**0.0002**
	Ctrl	82	1.0025	1.0400	1.0700	1.0348	0.0672	PE > 37 vs PE34-37	**0.0327**
								Ctrl vs PE34-37	**0.0000**
								Ctrl vs PE > 37	**0.0000**
UtA-PI MoM	PE < 34	11	0.9700	1.0900	1.3200	1.1555	0.2587	PE34-37 vs PE < 34	0.8605
	PE 34-37	11	0.9150	1.0300	1.2450	1.0836	0.2579	PE > 37 vs PE < 34	0.6412
	PE > 37	60	0.9150	1.0450	1.2125	1.0727	0.2381	Ctrl vs PE < 34	**0.0181**
	Ctrl	82	0.8400	0.9300	1.0700	0.9515	0.1811	PE > 37 vs PE34-37	0.9986
								Ctrl vs PE34-37	0.2229
								Ctrl vs PE > 37	**0.0059**
BHCG MoM	PE < 34	11	0.7785	0.9870	1.0595	1.0567	0.5621	PE34-37 vs PE < 34	0.9261
	PE 34-37	11	0.5300	1.0280	1.8960	1.2619	0.8744	PE > 37 vs PE < 34	0.8229
	PE > 37	60	0.6630	1.0440	1.4783	1.2775	0.9533	Ctrl vs PE < 34	0.9848
	Ctrl	82	0.7218	0.9470	1.4295	1.1451	0.6330	PE > 37 vs PE34-37	0.9999
								Ctrl vs PE34-37	0.9660
								Ctrl vs PE > 37	0.7487
PAPP A MoM	PE < 34	11	0.4900	0.8360	1.2840	0.8940	0.5445	PE34-37 vs PE < 34	0.3260
	PE 34-37	11	0.8895	1.2000	1.4005	1.1690	0.4536	PE > 37 vs PE < 34	0.1520
	PE > 37	60	0.7170	1.0440	1.4658	1.2717	0.9127	Ctrl vs PE < 34	**0.0081**
	Ctrl	82	0.9473	1.1755	1.7840	1.4263	0.7938	PE > 37 vs PE34-37	0.9997
								Ctrl vs PE34-37	0.7665
								Ctrl vs PE > 37	0.1856
PlGF MoM	PE < 34	11	0.8000	0.9570	1.1365	1.0577	0.5938	PE34-37 vs PE < 34	0.9997
	PE 34-37	11	0.6980	1.0860	1.2685	1.0188	0.4585	PE > 37 vs PE < 34	0.3054
	PE > 37	60	0.9265	1.3760	1.6105	1.3681	0.6508	Ctrl vs PE < 34	0.0578
	Ctrl	82	1.0455	1.2840	1.8893	1.4942	0.6548	PE > 37 vs PE34-37	0.2493
								Ctrl vs PE34-37	**0.0424**
								Ctrl vs PE > 37	0.4794

detection rates for the previously mentioned groups were 50% (30/60), 48.3% (29/60) and 51.7%, (31/60), respectively.

5. Discussion

5.1. The importance of population-specific maternal characteristics and biophysical measurements

According to our knowledge, this is the first prospective study performed in Hungary with their characteristic a priori risk and biophysical measurements. We calculated a priori risks based on biophysical and biochemical measurements for PE using patient cohorts from the United Kingdom, with their higher cardiovascular risk factors and the high percentage of patients of Afro-Caribbean origin (Yu et al., 2006). Previous studies have highlighted that maternal risk factors for PE might vary considerably in other populations (Shamsi et al., 2010; Lobo et al., 2019), which necessitates adjustment of the a priori risks and biophysical measurements in each population.

5.2. Comparable data with studies of non-Anglo-Saxon populations

Early-PE is the less frequent form of PE but is the major contributor to maternal and perinatal morbidity and mortality. This study demonstrated that screening for early-PE could be achieved in routine clinical practice in Eastern Europe at a DR of 72.7% when using maternal characteristics, biophysical measurements, and PAPP-A. Compared with the DRs of the FMF (89.2%) (Poon et al., 2009c), our DRs are lower and similar to some other studies (Oliveira et al., 2014; Teixeira

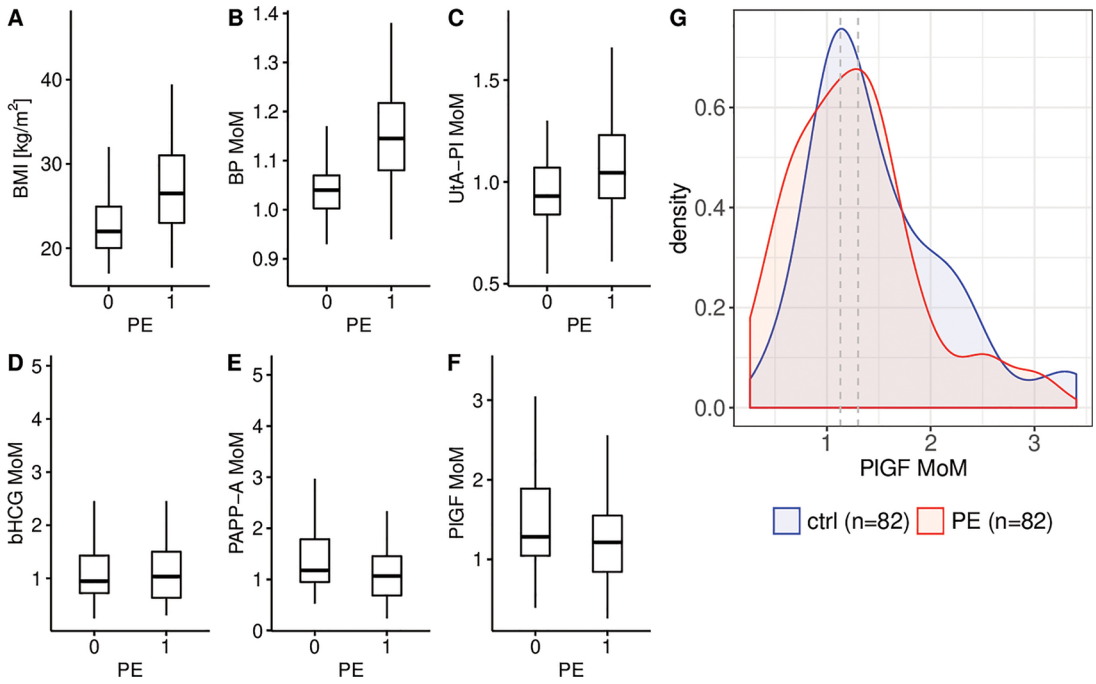

Fig. 1. Pregnancy characteristics in PE compared with control group illustrated with box plots. 0 — control group (n = 82), 1 — PE group (n = 82), and the distributions of PlGF MoMs in the control and PE populations. Blood pressure (BP), multiple of expected median (MoM), uterine artery pulsatility index (UtA-PI), beta human choriogonadotropin (bHCG), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF).

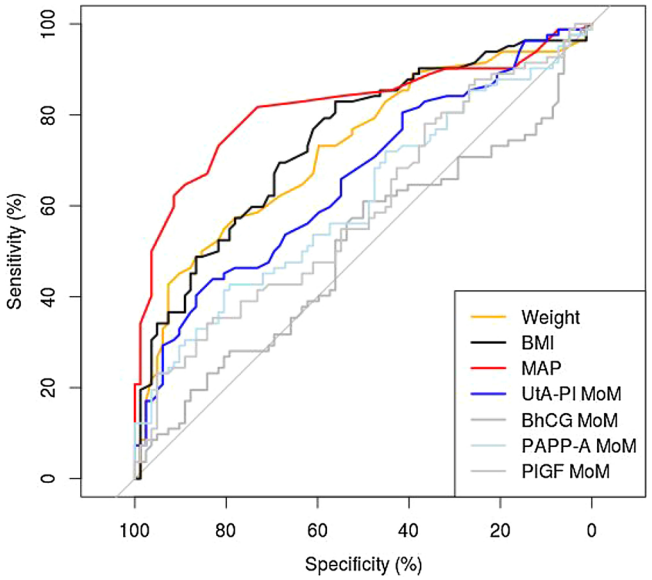

Fig. 2. Receiver–operating characteristics curves for prediction of PE. Body mass index (BMI), mean arterial pressure (MAP), uterine artery pulsatility index (UtA-PI), multiple of expected median (MoM), beta human choriogonadotropin (BhCG), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF).

et al., 2014; Cheng et al., 2018; Sepúlveda-Martínez et al., 2019) performed under routine clinical practice rather than in research settings. Apart from performing this study under routine clinical practice, another explanation for the lower DRs is the relatively low number of early-PE cases. Chronic hypertension and previous history of PE have a high influence on the risk assessment for early-PE. In our study among the 11 early-PE cases, there were only one with chronic hypertension and none with previous history of PE. Further Hungarian studies with larger patient numbers are needed to refine the early-PE patient characteristics. The detection rates for early-PE with A2 software was higher than with A1 software probably due to the improvement in screening algorithms. A2 software is recommended for use in routine clinical practice. The detection rate for late-PE in our study was comparable with the literature data. The reason why at the moment it is hardly feasible to enhance the detection rate of late-PE is due to the features of the disease (Lampe et al., 2011; Haram et al., 2014; Hahn et al., 2015; Erez et al., 2017; Lampe et al., 2017). Late-PE is mostly influenced by maternal factors rather than placental disease, keeping it challenging to predict and highlighting the importance of the a priori risk and biophysical measurements (Crispi et al., 2006).

5.3. The role of biochemical measurements

5.3.1. BhCG and PAPP-A

Our study is the first prospective assessment of PE screening in Hungary incorporating individual risk factors and biophysical measurements which are different from those used in other studies with higher DRs in the United Kingdom (Wright et al., 2019a). BP MoM, BMI, and UtA-PI Doppler were the strongest predictors of early-PE in this study, which is consistent with the results of Poon et al. (2009b). Similarly to other studies, BhCG did not show significant differences between the affected and the healthy groups (Abdi et al., 2018). Regarding PAPP-A, previous studies suggested a close association between low PAPP-A and PE (Pilalis et al., 2007), but the recent studies by FMF showed that in combination with other markers it becomes insignificant (Akolekar et al., 2009). PAPP-A proved to be an essential marker of the most common trisomies in the first-trimester screening, and in our study PAPP-A measurement seemed to be a useful tool for the improvement of the detection rate of PE as well. At the same time, the FPR decreased from 2.4% (2/82) to 0% (0/82) in our control group when we incorporated PAPP-A results into the risk calculation. However, this finding might be accidental because of the low number of control patients. It seems to be worthy of investigating the role of PAPP-A in the improvement of the FPR. When no other biomarkers are measured in routine clinical practice, but PAPP-A levels are available from common trisomy risk calculation, the result could also be useful for the risk calculation of PE.

Table 5
Detection rates of A1 and A2 softwares in different settings. Preeclampsia (PE), placenta associated plasma protein A (PAPP-A), placental growth factor (PlGF).

PE	Astraia 2.3.2 (A1) without biochemistry	Astraia 2.3.2 (A1) PAPP-A	Astraia 2.8.1(A2) without biochemistry	Astraia 2.8.1 (A2) PAPP-A	Astraia 2.8.1 (A2) PAPP-A + PlGF
Early-PE X < 34 wks (n = 11)	63.6% (7/11)	54.5% (6/11)	63.6% (7/11)	72.7% (8/11)	45.5% (5/11)
PE 34 > X > 37 wks (n = 11)	–	–	90.1% (10/11)	90.1% (10/11)	72.7% (8/11)
PE X > 37 wks (n = 60)	–	–	50% (30/60) (2FPR)	48.3% (29/60) (0FPR)	51.7% (31/60) (4FPR)
Late-PE x > 34 wks (n = 71)	67.6% (48/71) (5FPR)	64.8% (46/71) (5FPR)	56.3% (40/71) (2FPR)	54.9% (39/71) (0FPR)	54.9% (39/71) (4FPR)

5.3.2. PlGF

The use of PlGF not only did not increase but, in contrary to other studies, even deteriorated the detection rates. It is most likely due to the high mean levels of PlGF MoMs both in the PE and the control groups (1.215 MoM vs. 1.284 MoM). The FMF algorithm converts all measurements into MoMs, but the calculation uses data drawn from the Anglo-Saxon population. It is of a high probability that the distribution of PlGF levels in pregnancies both with the normal outcome and PE shows a different pattern in the Eastern European population due to several factors. For this reason, it is of utmost importance to prepare characteristic curves for the Hungarian population based on much more data. Mean PlGF levels in our study as demonstrated in Tables 4a and 4b were significantly lower in pregnancies that ended up with preeclampsia between 34–37 weeks (1.086 MoM) when compared to the controls (1.284 MoM), and the difference was almost significant between early-PE group (0.96 MoM) vs. control group as well. This finding is consistent with the literature data (Kusanovic et al., 2009). We believe that the detection rate of the screening for early preeclampsia could be significantly improved if one would adjust converted MoMs in the PE risk calculation algorithm to the population-specific Hungarian curve. Nevertheless, the results of our study together with several others necessitates the search for further potentially more effective biochemical markers (Erez et al., 2008; Romero et al., 2008; Than et al., 2008; Romero et al., 2017; Than et al., 2018; Rafaeli-Yehudai et al., 2018).

5.4. Possible cost-effective strategies of PE screening in Eastern Europe

From a financial point of view, in the Eastern part of Europe, the socioeconomic status of the population is much lower than in Western European countries. Most of the population use the national health care system where the combined test is not supported financially and is only available in the private healthcare sector, meanwhile, the routine first-trimester ultrasound is part of the national health care system. Using the latest software (A2) without biochemical markers, the detection rate for early-PE and late-PE was 63.6% (7/11) and 56.3% (40/71), respectively. It highlights that in routine clinical practice the maternal a priori risk combined with BP and UtA-PI measurements performed both by FMF standards can provide an acceptable screening method for early-PE for those who cannot afford the cost of the combined test. With this approach, the detection rate for early-PE without the biochemical markers is slightly under the detection rate of research studies using the same algorithm and presented by the FMF (Poon et al., 2009a). The study by Poon et al. (2009b) also showed that adding UtA-PI to the maternal factors (a priori risk) the detection rate could be raised from 31% to 73%. The International Society for the Study of Hypertension in Pregnancy (ISSHP) supports first trimester screening for risk of PE when this can be integrated into the local health system, although the cost effectiveness of this approach remains to be established (Brown et al., 2018).

5.5. Ethical issues — counselling before screening for early-PE and counseling high risk patients

Each patient participating in screening for PE in the first trimester of pregnancy needs to be informed on the efficacy of screening and also on the FPR. In case of a high risk result, the patient has to be reassured that according to our recent knowledge acetylsalicylic acid has no teratogenic effect nor it can cause any significant adverse effects in pregnancy such as placental abruption or vaginal bleeding when given in an appropriate dose and time (Bujold et al., 2009; Roberge et al., 2018). The recommended dose is between 100–150 mg/day, until the 34th weeks of gestation (Wright and Nicolaides, 2019b). In order to lower the anxiety of screen positive patients for early-PE, their first trimester risks can be recalculated at the 18–22 weeks scan (Wright et al., 2019a).

6. Conclusion

Compared with other papers, this study also supports the evidence that first-trimester screening for PE is feasible. Even more, this screening method could be integrated into the routine clinical first-trimester screening, when apart from screening for fetal malformations and chromosomal abnormalities for a minimal extra cost PE screening could also be performed. The accuracy of maternal and fetal biophysical measurements has a significant impact on the efficacy of PE screening. It highlights the need for standardized measurements on UtA-PI, CRL, and BP as recommended by FMF guidelines and measurement protocols. It can be expected that the prophylactic treatment with aspirin started in the first trimester in the high-risk group will result in a drop of PE prevalence at a minimal extra cost (Roberge et al., 2017; Wright et al., 2018). Future studies are required with a higher number of patients to validate the normal and abnormal distribution of PlGF in Eastern European population. Those may contribute to the improvement of the screening efficacy in the Eastern European countries. Our study also points out the need for further studies aiming the search for more effective biomarkers in preeclampsia screening.

Competing interest

The authors declare that they have no competing interest.

Author contributions

OL, OT, ZT conceptualized the original prospective study and designed research. OL, OT, NGT, RP designed the present cohort study, OL, OT, NGT analyzed and interpreted data OL, OG, OL Sr, AF, LM, KP recruited the patients and performed the screening, LV performed the biochemical measurements, OL, OG, DD, IA collected the follow up data and the risk calculations, ZsK performed the statistical analyses. All authors contributed to manuscript writing and approved the paper.

Declaration of interest

None.

Funding

This work was supported by grants from the Hungarian Academy of Sciences (Momentum LP2014-7/2014) and from the Hungarian National Research, Development and Innovation Fund (FIEK_16-1-2016-0005). The funding sources had no role in the design and execution of this study, analyses and interpretation of the data, or decision to submit results.

Acknowledgements

We thank Angela Molnarne Ori, Franciska Debreceni, Szilvia Nagy, Istvanne Sos, Erika Magyar, Csilla Kissne (Ultrasound Department, University of Debrecen Medical and Health Science Centre), Ildiko Veresne, Ildiko Medvene Jaszai, (Ultrasound Department, Andras Josa County and Teaching Hospital) and Dr. Csaba Karabelyos (Biotest Hungaria LTD) for their assistance.

References

Abdi, F., Aghaie, Z., Rahnemaie, F.S., Alimoradi, Z., 2018. A systematic review of first trimester biochemical and molecular predictive tests for preeclampsia. Curr. Hypertens. Rev. 14 (1), 21–28. https://doi.org/10.2174/1573402114666180416160425.

Agarwal, R., Chaudhary, S., Kar, R., Radhakrishnan, G., Tandon, A., 2017. Prediction of preeclampsia in primigravida in late first trimester using serum placental growth factor alone and by combination model. J. Obstet. Gynaecol. (Lahore) 37 (7), 877–882. https://doi.org/10.1080/01443615.2017.1309367.

Akolekar, R., Minekawa, R., Veduta, A., Romero, X.C., Nicolaides, K.H., 2009. Maternal plasma inhibin A at 11-13 weeks of gestation in hypertensive disorders of pregnancy. Prenat. Diagn. 29 (8), 753–760. https://doi.org/10.1002/pd.2279.

Bahado-Singh, R.O., Syngelaki, A., Akolekar, R., Mandal, R., Bjondahl, T.C., Han, B., Dong, E., Bauer, S., Alpay-Savasan, Z., Graham, S., Turkoglu, O., Wishart, D.S., Nicolaides, K.H., 2015. Validation of metabolomic models for prediction of early-onset preeclampsia. Am. J. Obstet. Gynecol. 213 (4). https://doi.org/10.1016/j.ajog.2015.06.044. 530.e1-530.e10.

Baschat, A., Magder, L., Doyle, L., Atlas, R., Jenkins, C., Blitzer, M., 2014. Prediction of preeclampsia utilizing the first trimester screening examination. Am. J. Obstet. Gynecol. 211 (5). https://doi.org/10.1016/j.ajog.2014.04.018. 514.e1–7.

Brown, M., Magee, L., Kenny, L., Karumanchi, S., McCarthy, F., Saito, S., Hall, D., Warren, C., Adoyi, G., Ishaku, S., 2018. International Society for the Study of Hypertension in Pregnancy (ISSHP)., 2018. The hypertensive disorders of pregnancy: ISSHP classification, diagnosis & management recommendations for international practice. Pregnancy Hypertens. 13, 291–310. https://doi.org/10.1016/j.preghy.2018.05.004.

Bujold, E., Morency, A., Roberge, S., Lacasse, Y., Forest, J., Giguère, Y., 2009. Acetylsalicylic acid for the prevention of preeclampsia and intra-uterine growth restriction in women with abnormal uterine artery Doppler: a systematic review and meta-analysis. J. Obstet. Gynaecol. Can. 31 (September (9)), 818–826. https://doi.org/10.1016/S1701-2163(16)34300-6.

Chaiworapongsa, T., Chaemsaithong, P., Yeo, L., Romero, R., 2014a. Pre-eclampsia part 1: current understanding of its pathophysiology. Nat. Rev. Nephrol. 10 (8), 466–480. https://doi.org/10.1038/nrneph.2014.102.

Chaiworapongsa, T., Chaemsaithong, P., Korzeniewski, S.J., Yeo, L., Romero, R., 2014b. Pre-eclampsia part 2: prediction, prevention and management. Nat. Rev. Nephrol. 10 (9), 531–540. https://doi.org/10.1038/nrneph.2014.103.

Cheng, Y., Leung, T.Y., Law, L.W., Ting, Y.H., Law, K.M., Sahota, D.S., 2018. First trimester screening for pre-eclampsia in Chinese pregnancies: case-control study. BJOG 125 (4), 442–449. https://doi.org/10.1111/1471-0528.14970.

Crispi, F., Domínguez, C., Llurba, E., Martín-Gallan, P., Cabero, L., Gratacos, E., 2006. Placental angiogenic growth factors and uterine artery Doppler findings for characterization of different subsets in preeclampsia and in isolated intrauterine growth restriction. Am. J. Obstet. Gynecol. 195 (1), 201–207.

Dhariwal, N.K., Lynde, G.C., 2017. Update in the management of patients with pre-eclampsia. Anesthesiol Clin. Mar 35 (1), 95–106. https://doi.org/10.1016/j.anclin.2016.09.009.

Duley, L., 2009. The global impact of pre-eclampsia and eclampsia. Semin. Perinatol. 33 (3), 130–137. https://doi.org/10.1053/j.semperi.2009.02.010.

Erez, O., Romero, R., Espinoza, J., Fu, W., Todem, D., Kusanovic, J.P., Gotsch, F., Edwin, S., Nien, J.K., Chaiworapongsa, T., Mittal, P., Mazaki-Tovi, S., Than, N.G., Gomez, R., Hassan, S.S., 2008. The change in concentrations of angiogenic and anti-angiogenic factors in maternal plasma between the first and second trimesters in risk assessment for the subsequent development of preeclampsia and small-for-gestational age. J. Matern. Fetal. Neonatal. Med. 21 (5), 279–287. https://doi.org/10.1080/14767050802034545.

Erez, O., Romero, R., Maymon, E., Chaemsaithong, P., Done, B., Pacora, P., Panaitescu, B., Chaiworapongsa, T., Hassan, S.S., Tarca, A.L., 2017. The prediction of late-onset preeclampsia: results from a longitudinal proteomics study. PLoS One 12 (7), e0181468. https://doi.org/10.1371/journal.pone.0181468.

Foidart, J.M., Munaut, C., Chantraine, F., Akolekar, R., Nicolaides, K.H., 2010. Maternal plasma soluble endoglin at 11-13 weeks' gestation in pre-eclampsia. Ultrasound Obstet. Gynecol. 35 (6), 680–687. https://doi.org/10.1002/uog.7621.

Gallo, D., Poon, L.C., Fernandez, M., Wright, D., Nicolaides, K.H., 2014. Prediction of preeclampsia by mean arterial pressure at 11-13 and 20-24 weeks' gestation. Fetal. Diagn. Ther. 36 (1), 28–37. https://doi.org/10.1159/000360287.

Gardosi, J., Kady, S.M., McGeown, P., Francis, A., Tonks, A., 2005. Classification of stillbirth by relevant condition at death (ReCoDe): population-based cohort study. BMJ 331 (7525), 1113–1117.

Hahn, S., Lapaire, O., Than, N.G., 2015. Biomarker development for presymptomatic molecular diagnosis of preeclampsia: feasible, useful or even unnecessary? Expert Rev. Mol. Diagn. 15 (5), 617–629. https://doi.org/10.1586/14737159.2015.1025757.

Haram, K., Mortensen, J.H., Nagy, B., 2014. Genetic aspects of preeclampsia and the HELLP syndrome. J. Pregnancy 2014, 910751. https://doi.org/10.1155/2014/910751.

Khalil, A., Garcia-Mandujano, R., Maiz, N., Elkhouli, M., Nicolaides, K.H., 2014. Longitudinal changes in uterine artery Doppler and blood pressure and risk of pre-eclampsia. Ultrasound Obstet. Gynecol. 43 (5), 541–547. https://doi.org/10.1002/uog.13257.

Kusanovic, J.P., Romero, R., Chaiworapongsa, T., Erez, O., Mittal, P., Vaisbuch, E., Mazaki-Tovi, S., Gotsch, F., Edwin, S.S., Gomez, R., Yeo, L., Conde-Agudelo, A., Hassan, S.S., 2009. A prospective cohort study of the value of maternal plasma concentrations of angiogenic and anti-angiogenic factors in early pregnancy and midtrimester in the identification of patients destined to develop preeclampsia. J. Matern. Fetal. Neonatal. Med. 22 (11), 1021–1038. https://doi.org/10.3109/14767050902994754.

Lampe, R., Szucs, S., Adany, R., Poka, R., 2011. Granulocyte superoxide anion production and regulation by plasma factors in normal and preeclamptic pregnancy. J. Reprod. Immunol. 89 (2), 199–206. https://doi.org/10.1016/j.jri.2011.01.019.

Lampe, R., Kover, A., Szucs, S., Pal, L.A., Poka, R., 2017. The effect of healthy pregnant plasma and preeclamptic plasma on the phagocytosis index of neutrophil granulocytes and monocytes of nonpregnant women. Hypertens. Pregnancy 36 (1), 59–63. https://doi.org/10.1080/10641955.2016.1237644.

Lobo, G.A.R., Nowak, P.M., Panigassi, A.P., Lima, A.I.F., Araujo Junior, E., Nardozza, L.M.M., Pares, D.B.S., 2019. Validation of Fetal Medicine Foundation algorithm for prediction of pre-eclampsia in the first trimester in an unselected Brazilian population. J. Matern. Fetal. Neonatal. Med. 32 (2), 286–292. https://doi.org/10.1080/14767058.2017.1378332.

Mayrink, J., Costa, M.L., Cecatti, J.G., 2018. Preeclampsia in 2018: revisiting concepts, physiopathology, and prediction. Scientific World Journal 2018, 6268276. https://doi.org/10.1155/2018/6268276.

O'Gorman, N., Wright, D., Poon, L.C., Rolnik, D.L., Syngelaki, A., de Alvarado, M., Carbone, I.F., Dutemeyer, V., Fiolna, M., Frick, A., Karagiotis, N., Mastrodima, S., de Paco Matallana, C., Papaioannou, G., Pazos, A., Plasencia, W., Nicolaides, K.H., 2017. Multicenter screening for preeclampsia by maternal factors and biomarkers at 11-13 weeks' gestation: comparison to NICE guidelines and ACOG recommendations. Ultrasound Obstet. Gynecol. 49 (6), 756–760. https://doi.org/10.1002/uog.17455.

Oliveira, N., Magder, L.S., Blitzer, M.G., Baschat, A.A., 2014. First-trimester prediction of pre-eclampsia: external validity of algorithms in a prospectively enrolled cohort. Ultrasound Obstet. Gynecol. 44 (3), 279–285. https://doi.org/10.1002/uog.13435.

Pilalis, A., Souka, A.P., Antsaklis, P., Daskalakis, G., Papantoniou, N., Mesogitis, S., Antsaklis, A., 2007. Screening for pre-eclampsia and fetal growth restriction by uterine artery Doppler and PAPP-A at 11-14 weeks' gestation. Ultrasound Obstet. Gynecol. 29 (2), 135–140.

Poon, L.C., Maiz, N., Valencia, C., Plasencia, W., Nicolaides, K.H., 2009a. First-trimester maternal serum pregnancy-associated plasma protein-A and pre-eclampsia. Ultrasound Obstet. Gynecol. 33 (1), 23–33. https://doi.org/10.1002/uog.6280.

Poon, L.C., Kametas, N.A., Maiz, N., Akolekar, R., Nicolaides, K.H., 2009b. First-trimester prediction of hypertensive disorders in pregnancy. Hypertension. 53 (5), 812–818. https://doi.org/10.1161/hypertensionaha.108.127977.

Poon, L.C., Staboulidou, I., Maiz, N., Plasencia, W., Nicolaides, K.H., 2009c. Hypertensive disorders in pregnancy: screening by uterine artery Doppler at 11-13 weeks. Ultrasound Obstet. Gynecol. 34 (2), 142–148. https://doi.org/10.1002/uog.6452.

Poon, L.C., Kametas, N.A., Chelemen, T., Leal, A., Nicolaides, K.H., 2010a. Maternal risk factors for hypertensive disorders in pregnancy: a multivariate approach. J. Hum. Hypertens. 24 (2), 104–110. https://doi.org/10.1038/jhh.2009.45.

Poon, L.C., Stratieva, V., Piras, S., Piri, S., Nicolaides, K.H., 2010b. Hypertensive disorders in pregnancy: combined screening by uterine artery Doppler, blood pressure and serum PAPP-A at 11-13 weeks. Prenat. Diagn. 30 (3), 216–223. https://doi.org/10.1002/pd.2440.

R Core Team, 2018. R: a Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. URL: https://www.R-project.org/. (2019.01.10.). .

Rafaeli-Yehudai, T., Imterat, M., Douvdevani, A., Tirosh, D., Benshalom-Tirosh, N., Mastrolia, S.A., Beer-Weisel, R., Klaitman, V., Riff, R., Greenbaum, S., Alioshin, A., Rodavsky Hanegbi, G., Loverro, G., Catalano, M.R., Erez, O., 2018. Maternal total cell-free DNA in preeclampsia and fetal growth restriction: Evidence of differences in maternal response to abnormal implantation. PLoS One 13 (7), e0200360. https://doi.org/10.1371/journal.pone.0200360.

Roberge, S., Nicolaides, K.H., Demers, S., Hyett, J., Chaillet, N., Bujold, E., 2017. The role of aspirin dose on the prevention of preeclampsia and fetal growth restriction: systematic review and meta-analysis. Am. J. Obstet. Gynecol. 216 (2), 110–120.e6. https://doi.org/10.1016/j.ajog.2016.09.076.

Roberge, S., Bujold, E., Nicolaides, K., 2018. Meta-analysis on the effect of aspirin use for prevention of preeclampsia on placental abruption and antepartum hemorrhage. Am. J. Obstet. Gynecol. 218 (5), 483–489. https://doi.org/10.1016/j.ajog.2017.12.238.

Robinson, H.P., Sweet, E.M., Adam, A.H., 1979. The accuracy of radiological estimates of

gestational age using early fetal crown-rump length measurements by ultrasound as a basis for comparison. Br. J. Obstet. Gynaecol. 86 (7), 525–528.

Rolnik, D.L., Wright, D., Poon, L.C., O'Gorman, N., Syngelaki, A., de Paco Matallana, C., Akolekar, R., Cicero, S., Janga, D., Singh, M., Molina, F.S., Persico, N., Jani, J.C., Plasencia, W., Papaioannou, G., Tenenbaum-Gavish, K., Meiri, H., Gizurarson, S., Maclagan, K., Nicolaides, K.H., 2017a. Aspirin versus placebo in pregnancies at high risk for preterm preeclampsia. N. Engl. J. Med. 377 (7), 613–622. https://doi.org/10.1056/NEJMoa1704559.

Rolnik, D.L., Wright, D., Poon, L.C.Y., Syngelaki, A., O'Gorman, N., de Paco Matallana, C., Akolekar, R., Cicero, S., Janga, D., Singh, M., Molina, F.S., Persico, N., Jani, J.C., Plasencia, W., Papaioannou, G., Tenenbaum-Gavish, K., Nicolaides, K.H., 2017b. ASPRE trial: performance of screening for preterm pre-eclampsia. Ultrasound Obstet. Gynecol. 50 (4), 492–495. https://doi.org/10.1002/uog.18816.

Romero, R., Kusanovic, J.P., Than, N.G., Erez, O., Gotsch, F., Espinoza, J., Edwin, S., Chefetz, I., Gomez, R., Nien, J.K., Sammar, M., Pineles, B., Hassan, S.S., Meiri, H., Tal, Y., Kuhnreich, I., Papp, Z., Cuckle, H.S., 2008. First-trimester maternal serum PP13 in the risk assessment for preeclampsia. Am. J. Obstet. Gynecol. 199 (2), 122.e1–122.e11. https://doi.org/10.1016/j.ajog.2008.01.013.

Romero, R., Erez, O., Hüttemann, M., Maymon, E., Panaitescu, B., Conde-Agudelo, A., Pacora, P., Yoon, B.H., Grossman, L.I., 2017. Metformin, the aspirin of the 21st century: its role in gestational diabetes mellitus, prevention of preeclampsia and cancer, and the promotion of longevity. Am. J. Obstet. Gynecol. 217 (3), 282–302. https://doi.org/10.1016/j.ajog.2017.06.003.

Scazzocchio, E., Figueras, F., Crispi, F., Meler, E., Masoller, N., Mula, R., Gratacos, E., 2013. Performance of a first-trimester screening of preeclampsia in a routine care low-risk setting. Am. J. Obstet. Gynecol. 208 (3), 203.e1–203.e10. https://doi.org/10.1016/j.ajog.2012.12.016.

Sepúlveda-Martínez, A., Rencoret, G., Silva, M.C., Ahumada, P., Pedraza, D., Muñoz, H., Valdés, E., Parra-Cordero, M., 2019. First trimester screening for preterm and term pre-eclampsia by maternal characteristics and biophysical markers in a low-risk population. J. Obstet. Gynaecol. Res. 45 (1), 104–112. https://doi.org/10.1111/jog.13809.

Shamsi, U., Hatcher, J., Shamsi, A., Zuberi, N., Qadri, Z., Saleem, S., 2010. A multicenter matched case control study of risk factors for preeclampsia in healthy women in Pakistan. BMC Womens Health 10, 14. https://doi.org/10.1186/1472-6874-10-14.

Teixeira, C., Tejera, E., Martins, H., Pereira, A.T., Costa-Pereira, A., Rebelo, I., 2014. First trimester aneuploidy screening program for preeclampsia prediction in a portuguese obstetric population. Obstet. Gynecol. Int. 2014, 435037. https://doi.org/10.1155/2014/435037.

Than, N.G., Romero, R., Hillermann, R., Cozzi, V., Nie, G., Huppertz, B., 2008. Prediction of preeclampsia - a workshop report. Placenta 29 Suppl A, S83–5.

Than, N.G., Romero, R., Tarca, A.L., Kekesi, K.A., Xu, Y., Xu, Z., Juhasz, K., Bhatti, G., Leavitt, R.J., Gelencser, Z., Palhalmi, J., Chung, T.H., Gyorffy, B.A., Orosz, L., Demeter, A., Szecsi, A., Hunyadi-Gulyas, E., Darula, Z., Simor, A., Eder, K., Szabo, S., Topping, V., El-Azzamy, H., LaJeunesse, C., Balogh, A., Szalai, G., Land, S., Torok, O., Dong, Z., Kovalszky, I., Falus, A., Meiri, H., Draghici, S., Hassan, S.S., Chaiworapongsa, T., Krispin, M., Knöfler, M., Erez, O., Burton, G.J., Kim, C.J., Juhasz, G., Papp, Z., 2018. Integrated systems biology approach identifies novel maternal and placental pathways of preeclampsia. Front. Immunol. 8 (9), 1661. https://doi.org/10.3389/fimmu.2018.01661.

Timmis, A., Townsend, N., Gale, C., Grobbee, R., Maniadakis, N., Flather, M., Wilkins, E., Wright, L., Vos, R., Bax, J., Blum, M., Pinto, F., Vardas, P., ESC Scientific Document Group, 2017. European society of cardiology: cardiovascular disease statistics 2017. Eur. Heart J. 39 (7), 508–579. https://doi.org/10.1093/eurheartj/ehx628.

Tunstall-Pedoe, H., Kuulasmaa, K., Mahonen, M., Tolonen, H., Ruokokoski, E., Amouyel, P., 1999. Contribution of trends in survival and coronary-event rates to changes in coronary heart disease mortality: 10-year results from 37 WHO MONICA project populations; monitoring trends and determinants in cardiovascular disease. Lancet 353 (9164), 1547–1557.

Wright, D., Nicolaides, K.H., 2019b. Aspirin delays the development of preeclampsia. Am. J. Obstet. Gynecol. https://doi.org/10.1016/j.ajog.2019.02.034. pii: S0002-9378(19)30386-2.

Wright, D., Syngelaki, A., Akolekar, R., Poon, L.C., Nicolaides, K.H., 2015. Competing risks model in screening for preeclampsia by maternal characteristics and medical history. Am. J. Obstet. Gynecol. 213 (1), 62.e1–62.e10. https://doi.org/10.1016/j.ajog.2015.02.018.

Wright, D., Rolnik, D., Syngelaki, A., de Paco Matallana, C., Machuca, M., de Alvarado, M., Mastrodima, S., Tan, M., Shearing, S., Persico, N., Jani, J., Plasencia, W., Papaioannou, G., Molina, F., Poon, L., Nicolaides, K., 2018. Aspirin for Evidence-Based Preeclampsia Prevention trial: effect of aspirin on length of stay in the neonatal intensive care unit. Am. J. Obstet. Gynecol. 218 (6), 612.e1–612.e6. https://doi.org/10.1016/j.ajog.2018.02.014.

Wright, D., Tan, M., O'Gorman, N., Poon, L., Syngelaki, A., Wright, A., Nicolaides, K.H., 2019a. Predictive performance of the competing risk model in screening for pre-eclampsia. Am. J. Obstet. Gynecol. 220 (2), 199.e1–199.e13. https://doi.org/10.1016/j.ajog.2018.11.1087.

Yu, C.K., Smith, G.C., Papageorghiou, A.T., Cacho, A.M., Nicolaides, K.H., 2006. An integrated model for the prediction of pre-eclampsia using maternal factors and uterine artery Doppler velocimetry in unselected low-risk women. Am. J. Obstet. Gynecol. 195 (1), 330.