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Concomitant intrauterine growth restriction alters the lipoprotein profile in preeclampsia

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ABSTRACT

Objective: Preeclampsia and intrauterine growth restriction (IUGR) are related conditions. We aimed to characterise common lipid changes.

Methods: Triglyceride and cholesterol levels of patients 24–42 weeks of gestation with IUGR (n = 52), hypertensive IUGR (HIUGR, n = 28), and preeclampsia without IUGR (PE, n = 56) were compared to a control group (CTRL, n = 167). In addition, 60 sera (n = 10 of each pathology IUGR, HIUGR, PE (without IUGR) compared to n = 30 matched CTRL) of severe early onset cases < 34 weeks of gestation were chosen and further analysed by ultracentrifugation lipid subfractionation including VLDL, IDL, LDL, and HDL composition.

Results: In the full cohort we found low cholesterol in IUGR ($p = 0.0405$), while triglyceride levels were high in PE ($p < 0.0001$). Lipid concentrations in HIUGR did not differ significantly from CTRL. In the 60 patients analysed by lipid subfractionation, triglyceride levels were increased in the VLDL subfraction in PE ($p < 0.01$), however, LDL-bound ApoB and cholesterol levels were lower in IUGR and HIUGR ($p < 0.0001$ for total cholesterol and $p < 0.001$ for ApoB in both groups), but not in PE when compared to CTRL.

Conclusion: Low cholesterol, especially LDL cholesterol levels are a feature of IUGR while high triglyceride levels are a feature of preeclampsia. Increased VLDL-triglycerides suggest a disturbed conversion to LDL in preeclampsia. Of note, the presence of IUGR in hypertensive disorders further alters lipid profiles, which may explain heterogeneous data on lipid values for preeclampsia in the literature. Study groups have to be selected carefully to avoid misinterpretation.

1. Introduction

Preeclampsia frequently leads to maternal and perinatal morbidity or mortality and hence plays a pivotal role in obstetric care [1]. The pathogenesis still remains unclear. Current hypothesis states that circulating factors, released from the placenta, alter maternal endothelial function [2]. A key variable that may be equally important is the maternal susceptibility or overall sensitivity of the maternal endothelium to these circulating factors [3].

During normal pregnancy lipid levels increase dramatically in order to guarantee fetal and placental nutritional supply, and steroid hormone synthesis [4,5]. Several reports have suggested that women with preeclampsia display further changes in lipid metabolism. Overwhelming circulating levels of triglycerides as compared to normal hyperlipidemia in pregnancy have been widely reported. A recent meta-analysis of 74 studies found that preeclampsia was additionally associated with elevated total cholesterol and non-HDL-cholesterol in the third trimester [6]. However, data still are conflicting. Hentschke et al. [7] did not found any differences of neither triglyceride nor cholesterol levels between preeclampsia and uncomplicated pregnancies in their study cohort. The authors remarked, that changes in lipid metabolism may rather be an expression of placental insufficiency and diminished

Abbreviations: DSC, dextransulfate-cellulose; HDL, high-density lipoprotein; H.E.L.P., Heparin-mediated extracorporeal LDL-precipitation; HIUGR, preeclampsia with underlying IUGR; HL, hepatic lipase; HLT3, hyperlipoproteinemia type III; IDL, intermediate density lipoprotein; IUGR, intrauterine growth restriction; LDL, low density lipoprotein; LPL, lipoprotein lipase; PE, preeclampsia; PlGF, placental growth factor; sFlt, soluble fms-like tyrosine kinase; SGA, small for gestational age; VLDL, very low density lipoprotein

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foetal growth – a feature often associated with preeclampsia. Equally, own observations published 2003 [8] demonstrated lower cholesterol and apolipoprotein (apo)B concentration in the low-density lipoprotein (LDL) fractions. We suggested a disturbed conversion of very low-density lipoprotein (VLDL) to LDL in preeclampsia since apoB in the VLDL fraction was increased. The discrepancies to the recent meta-analysis are likely caused by differences in the definition of the study population. During the last decade it has become more and more evident that preeclampsia is a heterogeneous disorder that can be subgrouped according to clinical and pathophysiological features. The terms 'maternal' and 'placental' preeclampsia have been introduced to underline that different underlying conditions and mechanisms might result in a similar clinical manifestation. Maternal preeclampsia is attributed by large to maternal preconditions and usually occurs late or near term in pregnancy (late-onset preeclampsia). It may be often secondary to an underlying disease. Predisposing risk factors include cardiovascular (arterial hypertension) and endocrine disorders (obesity, diabetes mellitus) [1]. Placental preeclampsia manifests early in pregnancy, before 34th week of gestation and is often complicated by intrauterine growth restriction (IUGR) [9]. The latter has been demonstrated to be associated with low cholesterol and LDL-cholesterol levels at least in the absence of preeclampsia [10,11]. Considering the heterogeneity of preeclampsia subgroups and its association to IUGR we now aimed to extend the lipid analyses to gain a deeper understanding of lipid metabolism and its role in the pathogenesis of both diseases. We hypothesized that in general IUGR displays a low cholesterol phenotype while pure preeclampsia is associated with high triglyceride concentrations.

2. Materials and methods

2.1. Participants and study protocol

The study was approved by the Ethics Committee of the Medical Faculty of the Technical University RWTH Aachen (EK 138/06, 148/07, 119/08 and 154/11) and each patient gave written informed consent. Biomaterial was sampled from July 2006 to March 2012. In total 303 patients with complete antenatal lipid profiles (total cholesterol (TC) and triglyerides (TG)) with blood drawn and analysed between 24 and 42 weeks of gestation (WOG), and prior to the beginning of active labour were included. Patients were in part already described in "*The Evaluation of the Oxidative State of Low-Density Lipoproteins in Intrauterine Growth Restriction and Preeclampsia*" by Pecks et al. [10]. The analysed groups were defined according to the following criteria, as previously described [10]:

(1) IUGR without hypertension (n = 52)
(2) IUGR with hypertension/preeclampsia (HIUGR) (n = 28)
(3) Preeclampsia without IUGR/small for gestational age neonate (PE) (n = 56)
(4) Control group; gestational age matched uncomplicated pregnancies (CTRL) (n = 167)

Gestational age was calculated by the last menstrual period, verified by first trimester scan documentation. For sonographic examinations, the GE Logiq 5® or GE Voluson 730® Ultrasound System (Solingen, Germany) was used. Fetal weight was estimated by the regression equation including biparietal diameter, femur length, and head and abdominal circumferences, as proposed by Hadlock et al. [12]. Population-based newborn weight charts were used to determine fetal and neonatal birth weight [13].

IUGR was defined as an estimated fetal weight below the 10th percentile paralleled by one of the following criteria: (i) deceleration of fetal growth velocity during the last 4 weeks [14,15], (ii) elevated resistance index in umbilical artery Doppler sonography above 95th percentile or absent or reversed end-diastolic blood flow [16], (iii) fetal asymmetry (head to abdominal circumference ratio above 95th percentile) [17], or (iv) oligohydramnios (Amniotic Fluid Index < 5 cm) [14,18]. Neonatal weight was assessed for correct diagnosis of SGA < 10th percentile.

PE was defined according to the International Society for the Study of Hypertension in Pregnancy statement as new onset of hypertension > 140/90 mmHg and proteinuria more than 300 mg/d after 20th week of gestation. The criteria of preeclampsia were also fulfilled in case of hypertension with IUGR, and these patients were grouped in the HIUGR group [1].

In the CTRL group, neonatal weight was appropriate (within the 10th and 90th percentile) for gestational age, and delivery occurred at least at 37 0/7 weeks after an uneventful pregnancy.

Exclusion criteria were multiple gestation, fetal anomalies, abnormal fetal karyotype, patients with clinical or biochemical signs of infection, positive TORCH screening results, diabetes mellitus, gestational diabetes, or other severe metabolic disorders at time of sampling, and patient's withdrawal from the study and/or unavailability of maternal and fetal follow-up. None of the samples were obtained during labor.

60 sera (n = 10 in each pathology was matched for gestational age at sampling (± 7 days) with n = 30 CTRL) sampled at diagnosis of early-onset disease between 24 0/7 and 34 0/7 weeks of gestation from patients not in active labour at a non-defined fasting-state were processed as described previously [19], and used for extensive lipid subfractionation analysis. A power calculation has been performed based on the LDL-6-subfraction from Winkler et al. [8]. We choose a power (1 − β-error) of 80% and a level of significance (α-error) below 0.05 for each group and an allocation of 3 controls to 1 case. In this setting at least 9 cases and 25 controls had to be included.

2.2. Sequential ultracentrifugation

Serum was subjected to preparative sequential density ultracentrifugation with a target density < 1.006 kg/l for VLDL, < 1.019 kg/l for intermediate density lipoproteins (IDL), < 1.063 kg/l for LDL, and < HDL as previously described [8]. Total LDLs were separated into six classes with different densities according to Baumstark et al [20]. LDL-1, 1.019–1.031 kg/l; LDL-2, 1.031–1.034 kg/l; LDL-3, 1.034–1.037 kg/l; LDL-4, 1.037–1.040 kg/l; LDL-5, 1.040–1.044 kg/l; LDL-6, 1.044–1.063 kg/l. The operator was blinded to the status of the samples.

2.3. Determination of lipid parameters

ApoB, ApoA1, Cholesterol (total cholesterol, free cholesterol and cholesterolesters), triglycerides and phospholipids in serum and lipoproteinfractions were measured on an automated platform (Olympus AU 640) with commercially available test kits (DiaSys Greiner). Amounts of lipoproteins were given in mg/dL. The operator was blinded to the status of the samples.

2.4. Statistical analysis

GraphPad Prism V7.00 was used for statistical analysis. Data were tested for normal distribution with d'Agostino & Pearson normality test. Accordingly, statistical analysis was done with ANOVA or Kruskal-Wallis followed by Dunn's post-test. A value of $p < 0.05$ was considered to be statistically significant. Adjustment for BMI, gestational week at sampling, maternal age, smoking status, and parity was done by regression analysis and residual plot.

3. Results

3.1. Clinical characteristics

Table 1 shows the clinical characteristics of the study groups of the 303 patients. WOG at antenatal blood sampling was similar between

Table 1
Patient characteristics. Displayed are mean values and 95% CI. Statistical analysis was performed with one-way ANOVA. Significance for group effect is considered if p-value < 0.05.

		CTRL (n = 167)	IUGR (n = 52)	HIUGR (n = 28)	PE (n = 56)	p-value for group
Maternal age (y)	Mean	30.75	28.29	32.56	32.81	0.0006
	Lower 95% CI	29.9	26.74	30.1	31.25	
	Upper 95% CI	31.6	29.84	35.02	34.36	
BMI (kg/m^2)	Mean	24.3	23.67	27.77	26.51	< 0.0001
	Lower 95% CI	23.42	22.28	25.44	25.04	
	Upper 95% CI	25.19	25.07	30.09	27.97	
Primigravidity (%)	Mean	55.1	65.4	64.3	62.5	0.47
	Lower 95% CI	47.47	52.01	45.36	49.42	
	Upper 95% CI	62.71	78.76	83.21	75.58	
Smoking (%)	Mean	19.16	40.38	21.43	7.27	0.0004
	Lower 95% CI	13.13	26.59	5.23	0.19	
	Upper 95% CI	25.19	54.18	37.63	14.36	
Systolic blood pressure (mmHg)	Mean	115.9	117.8	150.4	157.8	< 0.0001
	Lower 95% CI	113.9	113.9	146.3	153	
	Upper 95% CI	118	121.8	154.4	162.6	
Diastolic blood pressure (mmHg)	Mean	67.03	68.62	92.07	94.8	< 0.0001
	Lower 95% CI	65.53	65.78	87.99	91.62	
	Upper 95% CI	68.53	71.45	96.15	97.98	
Gestational age at sampling (weeks)	Mean	34.09	31.75	30.85	33.56	0.0003
	Lower 95% CI	33.3	30.6	29.03	32.48	
	Upper 95% CI	34.87	32.91	32.67	34.64	
Gestational age at delivery (weeks)	Mean	38.95	33.3	32.63	34.43	< 0.0001
	Lower 95% CI	38.77	32.18	30.77	33.39	
	Upper 95% CI	39.12	34.42	34.48	35.46	
Neonatal weight (g)	Mean	3179	1470	1438	2233	< 0.0001
	Lower 95% CI	3102	1295	1158	1995	
	Upper 95% CI	3257	1646	1718	2472	

the study groups. Mean values differed in most of the variables including maternal BMI (with a significant higher BMI in the HIUGR and at least a borderline trend to increased BMI values in the PE group as compared to CTRL, $p < 0.0001$) and smoking status (with smokers were found most frequently in the IUGR group, $p = 0.0004$). Per definition blood pressure values were higher in PE and HIUGR groups ($p < 0.0001$), fetal weight was lower in the IUGR and in the HIUGR group as compared to CTRL ($p < 0.0001$). Table 2 similarly demonstrates the demographical data of the 60 patients used for detailed serum lipid subfractionation.

3.2. Serum lipid parameters

In the full cohort, serum triglycerides (TG) were significantly higher in the PE group than in the CTRL group ($p < 0.0001$). Total cholesterol (TC) levels were lower in the IUGR group than in the CTRL group ($p < 0.05$). Differences remained significant after adjustment for BMI, maternal age, gestational age at sampling, smoking status, and parity (table 3).

In the 60 patients used for extensive lipid analysis serum triglycerides were significantly higher in the PE group than in the CTRL group ($p < 0.05$). Total cholesterol ($p < 0.001$ for HIUGR and IUGR versus CTRL) and ApoB ($p < 0.001$ for IUGR and $p < 0.01$ for HIUGR versus CTRL) were lower in the IUGR and HIUGR group than in the CTRL or PE group. Phospholipids and ApoA1 were not different between the groups (Fig. 1). Metabolism of ApoB containing lipoproteins seems to be primarily affected and was therefore examined further.

3.3. Lipoprotein composition of the subgroup (n = 60)

Within the LDL fraction, all lipid components were significantly lower in the IUGR and HIUGR group than in the CTRL group (e. g. $p < 0.0001$ for total cholesterol, and $p < 0.001$ for ApoB in both, IUGR and HIUGR versus CTRL, respectively). In the VLDL fraction, triglyceride levels were higher in all three case groups compared to CTRL (table 4). The highest triglyceride level was found in the PE-group, being twofold higher than in the CTRL group ($p < 0.01$). In PE, the triglycerid-rich VLDL prevails, whereas in the IUGR state the LDL-fraction is reduced. Similar to the IUGR group, HIUGR has a reduced LDL fraction yet shows a tendency towards elevated VLDL and IDL as in PE, albeit not significant.

3.4. Lipoprotein profile of the subgroup (n = 60)

In the PE group ApoB-levels of the triglyceride-rich fraction VLDL are higher than in the CTRL group ($p < 0.01$), whereas in the IUGR group all levels of LDL-subfractions are lower than in the CTRL group (e.g. LDL-1 and LDL-2 $p < 0.05$). In the ApoB-profile the HIUGR group shows features of both the IUGR and the PE profiles, namely high triglyceride-rich lipoproteins (albeit not significant) and low LDL-fractions (e.g. LDL-1 $p < 0.01$ and LDL-2 $p < 0.001$) (Fig. 2).

4. Discussion

Changes in lipid concentrations in preeclampsia have been consistently reported. Yet, evidence is increasing that lipid metabolism depends on the type of preeclampsia and associated conditions. To gain deeper insights into changes involved in lipid metabolism and to highlight differences between subgroups we here analysed lipid levels in CTRL, PE without IUGR, and IUGR with and without hypertension in pregnancy. Consistent with the majority of reports of others (for review see: [21]) we found triglyceride levels to be higher in the PE group, yet LDL-particle composition was not affected in pure PE while LDL-particle number and -cholesterol content was reduced in IUGR as compared to the CTRL group irrespective of concomitant hypertension.

High triglycerides are a consequence of metabolic dysregulation due to enhanced VLDL-production, delayed hepatic remnant clearance or

Table 2
Patient characteristics of a subgroup of n = 30 severe early-onset pregnancy complications and n = 30 controls used for detailed lipid fractionation. Displayed are mean values and 95% CI. Statistical analysis was performed with one-way ANOVA. Significance for group effect is considered if p-value < 0.05.

		CTRL (n = 30)	IUGR (n = 10)	HIUGR (n = 10)	PE (n = 10)	p-value for group
Maternal age (y)	Mean	30.4	29.6	32.8	35.2	0.043
	Lower 95% CI	28.5	26.9	27.2	31.8	
	Upper 95% CI	32.4	32.3	38.3	38.6	
BMI (kg/m^2)	Mean	22.6	23.6	31.1	24.6	< 0.0001
	Lower 95% CI	21.5	20.7	27.0	22.6	
	Upper 95% CI	23.7	26.5	35.2	26.7	
Primigravidity (%)	Mean	60.0	80.0	90.0	40.0	0.082
	Lower 95% CI	41.4	49.8	67.4	3.1	
	Upper 95% CI	78.6	110.2	112.6	76.9	
Smoking (%)	Mean	6.7	40.0	20.0	0.0	0.028
	Lower 95% CI	−2.8	3.1	−10.2	0.0	
	Upper 95% CI	16.1	76.9	50.2	0.0	
Systolic blood pressure (mmHg)	Mean	116	125	150	162	< 0.0001
	Lower 95% CI	112	118	143	144	
	Upper 95% CI	121	132	158	180	
Diastolic blood pressure (mmHg)	Mean	69	72	95	99	< 0.0001
	Lower 95% CI	65	65	89	89	
	Upper 95% CI	73	79	100	108	
Gestational age at sampling (weeks)	Mean	29.0	29.1	28.4	30.7	0.173
	Lower 95% CI	28.0	27.4	26.1	28.9	
	Upper 95% CI	30.1	30.9	30.7	32.5	
Gestational age at delivery (weeks)	Mean	39.3	30.1	30.8	31.3	< 0.0001
	Lower 95% CI	38.7	28.4	27.4	29.5	
	Upper 95% CI	39.8	31.9	34.1	33.1	
Neonatal weight (g)	Mean	3339	945	1100	1549	< 0.0001
	Lower 95% CI	3156	678	676	1225	
	Upper 95% CI	3521	1212	1524	1873	
Neonatal weight percentiles	Mean	44.2	5.1	5.0	33.7	< 0.0001
	Lower 95% CI	35.2	3.0	2.6	18.3	
	Upper 95% CI	53.2	7.2	7.4	49.1	
Neonatal gender (% female)	Mean	53.3	50.0	50.0	60.0	0.967
	Lower 95% CI	34.4	12.3	12.3	23.1	
	Upper 95% CI	72.3	87.7	87.7	96.9	

disturbances in peripheral lipolysis [22]. These metabolic changes were by large regulated by specific enzymes like lipoprotein lipase (LPL), hepatic lipase (HL), cholesterolester transfer protein (CETP), and lecithin cholesterol acyltransferase (LCAT). Disturbances in enzyme activity are associated with distinct changes in lipid values. High triglyceride concentrations in the VLDL subfraction in preeclampsia suggest a disturbed conversion to LDL and a common enzymatic dysregulation at this step in the lipoprotein turnover. This lipid profile with high VLDL-triglycerides resembles an hyperlipidemia type III phenotype. Additionally, impaired conversion of large buoyant LDL and an increase in triglyceride-rich lipoproteins may be attributed to hepatic lipase (HL) deficiency [8].

By contrast to preeclampsia, lower cholesterol, especially LDL-cholesterol levels have been found in IUGR compared to CTRL. This is in line with previous reports [11,23]. Additionally, cholesterolester, phospholipids and ApoB in the LDL fraction were also significantly decreased, indicating both a reduced LDL-mass and particle number. This could support the notion that in IUGR, the mother's synthesis of VLDL – which deliver energy supply most efficiently via triglycerides – is maintained at the expense of IDL and LDL [11]. The cause of cholesterol-depletion in IUGR is not known by now. Insufficient stimulation of hepatic synthesis by reduced placental hormone synthesis may be one explanation [5]. Another explanation is an increased consumption and catabolism of LDL particles by the placenta. LDL-receptor overexpression and high oxidative stress levels with accumulation of oxidised LDL particles have been reported in IUGR placentae [24–27] suggesting an increased clearance of LDL particles from the maternal circulation.

In preeclampsia Mistry et al. observed an increased capacity of maternal blood to accept cholesterol from cells. The authors suggested a

Table 3
Lipid profile of 303 patients with complete lipid profile values. Significance was tested after adjustment for BMI, gestational week at sampling, maternal age, smoking status, and parity by Kruskal-Wallis test followed by Dunn's comparison test. Significant differences are marked in bold and by asterisk. *p < 0.05, **p < 0.01, ***p < 0.001.

		CTRL (n = 167)	IUGR (n = 52)	HIUGR (n = 28)	PE (n = 56)	p-value for group
TC mg/dL	Median	265	**238***	252	258	**0.0405**
	Lower 95% CI	255	214	205	233	
	Upper 95% CI	269	253	272	270	
TG mg/dL	Median	219	197	235	**298*****	**< 0.0001**
	Lower 95% CI	211	177	165	244	
	Upper 95% CI	228	214	271	333	

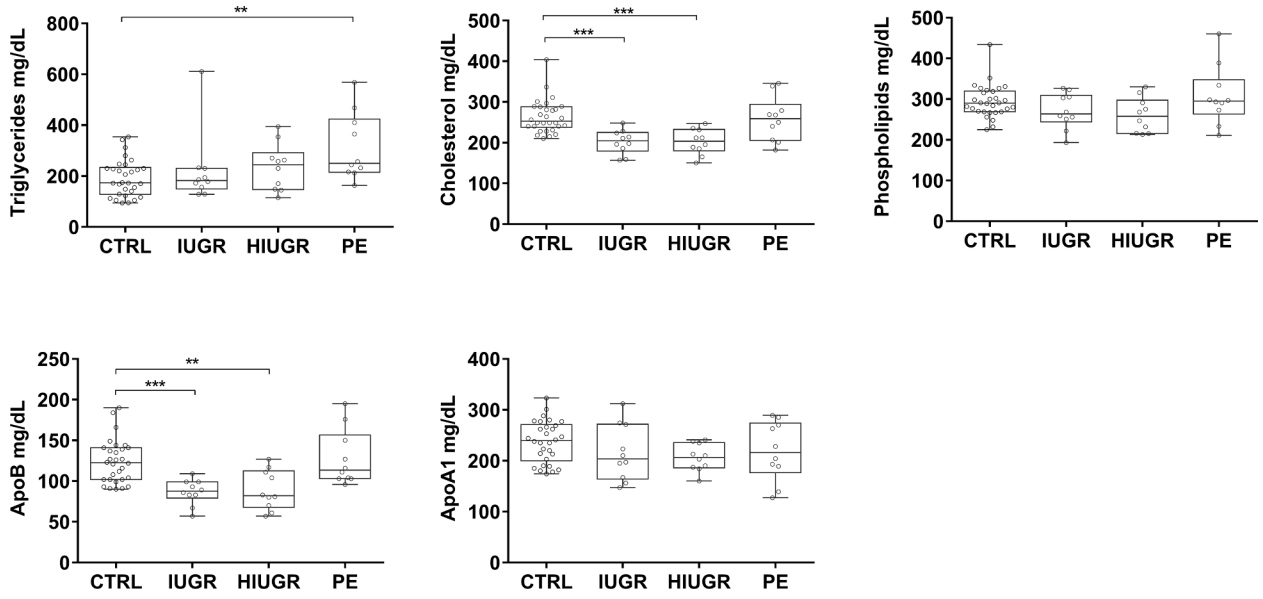

Fig. 1. Serum lipids in the subgroup of n = 30 severe early-onset pregnancy complications (n = 10 for IUGR, HIUGR and PE each) and n = 30 controls. Data are presented as box-plots (the box ranges from 25th to 75th percentile, the middle line represents the median, whiskers include all values from minimum to maximum and each value is represented by a point superimposed on the graph). Data were analysed with Kruskal-Wallis and Dunn's Post-test; *p < 0.05; **p < 0.01; ***p < 0.001; versus CTRL.

compensatory mechanism against cholesterol overload and lipid peroxidation in the placenta [28]. Like-wise, our own group demonstrated a lower cholesterol acceptor capacity in IUGR, at least in the foetal blood, which may exaggerate the accumulation of oxidised LDL particles in the placenta and reduce the overall cholesterol pool in the circulation [29]. It is accepted that IUGR and preeclampsia share common pathomechanisms. This is underlined by the fact that about 30% of patients with preeclampsia additionally have IUGR diagnosed and vice-versa. Oxidative stress and lipid peroxidation have been studied and discussed in the context of both diseases. However, distinct adaptive mechanisms take place which influence metabolism different. For example, LDL-receptor expression has been repeatedly shown to be increased in IUGR placentae [24–26] while in preeclampsia studies reported on no difference or even lower LDL-receptor expression when compared to normal pregnancies [7,30]. Moreover, even within a disease group, heterogeneous results have been published suggesting that

Table 4
Concentrations of lipid components in the lipid fractions. Data are presented as median (in bold numbers), lower 95% CI, upper 95% CI. Statistical analysis was performed with Kruskal-Wallis test followed by Dunn's post-test. Significant results are presented in italic. *p < 0.05; **p < 0.01; ***p < 0.001; ****p < 0.0001 versus CTRL.

		CTRL n = 30			IUGR (n = 10)			HIUGR (n = 10)			PE (n = 10)		
		Median	Lower 95% CI	Upper 95% CI	**Median**	Lower 95% CI	Upper 95% CI	**Median**	Lower 95% CI	Upper 95% CI	**Median**	Lower 95% CI	Upper 95% CI
VLDL	Total cholesterol	**24**	21	28	**24**	18	45	**37**	20	62	***39****	28	71
	Free cholesterol	**8**	6	10	**11**	6	19	**14**	7	24	***18****	11	33
	Cholesterolester	**16**	14	21	**16**	11	26	**23**	13	38	**20**	17	38
	Triglycerides	**97**	72	128	**120**	78	192	**148**	83	276	***178****	120	309
	Phospholipids	**30**	24	36	**37**	23	53	**47**	27	78	***55****	35	96
	Apolipoprotein B	**14**	13	17	**16**	11	25	**21**	15	33	***21****	17	42
IDL	Total cholesterol	**12**	10	17	**8**	6	15	**11**	8	17	**15**	9	22
	Free cholesterol	**3**	3	4	**2**	1	5	**3**	2	4	**4**	2	6
	Cholesterolester	**8,4**	7	12	**6**	4	11	**8**	6	13	**11**	6	16
	Triglycerides	**12**	11	15	**14**	9	20	**15**	12	20	**21**	12	35
	Phospholipids	**10**	9	13	**8**	6	16	**11**	8	16	**15**	8	22
	Apolipoprotein B	**8**	6	10	**7**	4	12	**8**	6	11	**10**	7	15
LDL	Total cholesterol	**126**	111	143	***75****	64	97	***82****	39	101	**117**	91	151
	Free cholesterol	**30**	28	36	***18****	14	25	***17****	8	24	**31**	22	36
	Cholesterolester	**95**	85	108	***58****	49	74	***62****	31	76	**89**	69	116
	Triglycerides	**34**	30	37	***24****	21	27	***25****	16	35	**35**	27	55
	Phospholipids	**85**	74	97	***54****	43	68	***56****	30	72	**86**	63	99
	Apolipoprotein B	**92**	78	99	***60****	52	71	***58****	31	79	**83**	63	103
HDL	Total cholesterol	**77**	73	81	**61**	50	97	***63****	50	72	**67**	43	78
	Free cholesterol	**14**	12	16	**10**	8	17	***10****	6	11	**11**	7	13
	Cholesterolester	**62**	61	65	**49**	41	81	***53****	41	61	**56**	35	65
	Triglycerides	**28**	25	33	***20****	13	28	**26**	15	37	**21**	16	35
	Phospholipids	**141**	121	153	**116**	105	190	**121**	103	146	**129**	76	146
	Apolipoprotein A1	**164**	150	182	**142**	122	195	**148**	132	175	**147**	111	170

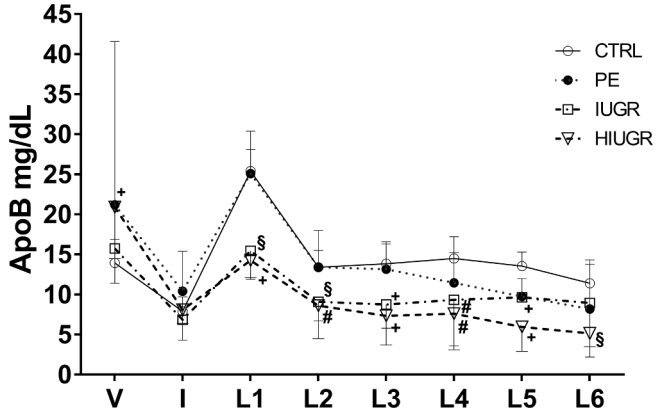

Fig. 2. ApoB values (in mg/dL) as median ± 95%CI in VLDL, IDL and the 6 different LDL-subclasses are shown. Data were analysed with Kruskal-Wallis followed by Dunn's Post-Test; § $p < 0.05$, + $p < 0.01$, # $p < 0.001$, vs. CTRL.

different pathomechanisms are involved leading to the same phenotype of IUGR and preeclampsia, respectively. For example, in different preeclampsia cohorts, some authors did report an increase in total cholesterol and/or LDL-cholesterol whereas others did not find differences between groups, or even report lower LDL-cholesterol [7,8,31,32] A large metaanalysis stated that LDL-cholesterol is higher in preeclampsia than in normal pregnancies [6], which cannot be supported by the results of the present study. By contrast, however, if additionally affected by IUGR quite the opposite is the case. As a main result of our present study it has therefore to be emphasised that preeclampsia is a heterogeneous disorder, and metabolic changes in preeclampsia are influenced by the placental function and the presence of IUGR.

It has been known for long that lipids are important risk factors in endothelial injury and cardiovascular disease [33,34] and may hence also contribute to endothelial dysfunction in preeclampsia [35]. During the last years interventional studies have been conducted or are ongoing for the prevention of preeclampsia or improvement of outcome using lipid modifying drug therapy [36]. Pravastatin, an inhibitor of the 3-hydroxy-3-methyl-glutaryl-coenzyme-A reductase (HMG-CoA-reductase) seemed to be promising, though it was assumed that beneficial effects are rather of pleiotropic nature than by its main mechanism of action, i.e. lowering LDL-cholesterol levels and restoring LDL/HDL ratio [37]. Unfortunately, in the recent "pravaStatin to Ameliorate early onset Pre-eclampsia (StAmP) trial" benefit, if any, was limited (personal communications Dr David Williams, London, GB at the EuroISSHP congress 2017). However, picking up the heterogenous data on lipid metabolism in preeclampsia, specific subtypes may indeed benefit from lipid modifying interventional strategies.

We recently demonstrated that Heparin-mediated extracorporeal LDL-precipitation (H.E.L.P.)-apheresis for the removal of atherogenic LDL-cholesterol showed promising results in prolonging preeclampsia pregnancies [38]. Potential mechanisms by which apheresis exerts its clinical benefit are controversially discussed [39,40] and ongoing studies will hopefully soon clarify the clinical benefit and mechanism of action. Given the varying clinical and biochemical phenotypes of preeclampsia, specifying the patient that can benefit most from these novel therapies appears to be crucial.

There are some limitations of the study. Potential and well known confounders of lipid values are BMI, gestational age at sampling, smoking or fasting status [6,21,41]. The HIUGR group had a higher BMI than the other groups, which might have influenced lipid values. A subgroup analysis within the group used for detailed lipid fractionation (n = 60) was not possible, as the number of patients would have been too small to perform meaningful statistical analyses. It is however remarkable, that the HIUGR group shows both features from IUGR and PE despite the higher BMI. Within the full cohort of n = 303, adjustment for BMI, maternal age, gestational age at sampling, smoking status, and parity did not majorly affected the results. The influence of BMI on our results seems therefore not to be a pivotal factor. Data from the full cohort and from the extensive analysis cohort differ in some aspects. In the full cohort TC is reduced only in IUGR, while TG are increased only in the PE group. The HIUGR cases showed only a tendency in lower TC (as observed in pure IUGR) and higher TG (as observed in pure PE). This inconsistency may arise from a sample bias because of limited sample number. Another explanation may be that the extensive analysis cohort consists only of severe early-onset cases, whereas the full cohort includes also late-onset cases, which may have differences in the pathophysiology. However, subcalculations of TG and TC concentrations separately for the early-onset groups and late-onset groups of the full cohort did show similar tendencies of biochemical markers in both cohorts (data not shown). Finally, a higher TG level and a lower TC level in the HIUGR group of the full cohort may be masked by lower concentrations of TG-containing LDL and higher concentration levels of cholesterol-containing VLDL. This underlines the importance of the extensive lipid analysis which has the potential to discover differences in subfractions.

5. Conclusion

Our results suggest a disturbed metabolism of ApoB containing lipoproteins, namely VLDL to LDL, in both IUGR and PE, with distinct differences depending on the underlying disease. In PE, triglyceride metabolism appears to be most affected whereas cholesterol metabolism is disturbed in IUGR irrespective of additional PE. Single studies about lipid metabolism in preeclampsia are controversial [32]. Our results suggest that this may be explained - at least in part - by one particular aspect of the multifaceted nature of preeclampsia, namely the presence or absence of IUGR.

This important factor therefore needs to be taken into account when interpreting lipid values in the context of preeclampsia, especially when planning interventional pharmacological studies.

6. Declarations of interest

None.

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