

# We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

## 4,800

Open access books available

## 122,000

International authors and editors

## 135M

Downloads

Our authors are among the

## 154

Countries delivered to

## TOP 1%

most cited scientists

## 12.2%

Contributors from top 500 universities

**WEB OF SCIENCE™**Selection of our books indexed in the Book Citation Index  
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?  
Contact [book.department@intechopen.com](mailto:book.department@intechopen.com)

Numbers displayed above are based on latest data collected.

For more information visit [www.intechopen.com](http://www.intechopen.com)

---

# Maternal Hypercholesterolemia in Gestational Diabetes and the Association with Placental Endothelial Dysfunction

A. Leiva, C Diez de Medina, E. Guzmán-Gutierrez,  
F. Pardo and L. Sobrevia

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/55297>

---

## 1. Introduction

Pregnancy is a physiological condition characterized by a progressive, weeks of gestation-dependent increase in maternal triglycerides (hypertriglyceridemia) and total cholesterol (hypercholesterolemia) [1-4]. In some cases a misadaptation occurs and these levels increase over a physiological range and dyslipidemia is recognized [5]. This condition occurs in some pregnancies coursing without associated pregnancy alterations [i.e., maternal supraphysiological hypercholesterolemia (MSPH)] and in pregnancies coursing with pathologies as preeclampsia and gestational diabetes mellitus (GDM) [3, 5].

GDM is widely associated with endothelial dysfunction of the placenta mainly triggered by hyperinsulinemia, hyperglycemia, and changes in nucleoside extracellular concentration and dyslipidemia associated with this pathology could play a role in this phenomenon since dyslipidemia is a risk factor to develop endothelial dysfunction and atherosclerosis [6]. Additionally, GDM predisposes to an accelerated development of cardiovascular disease (CVD) in adult life and as most of pregnancies with GDM course with elevated dyslipidemia, is feasible found a pathological link between dyslipidemia in GDM pregnancies and development of CVD later in life [6,7].

The hypertriglyceridemia described in GDM is directly related with the fetal macrosomia characteristic of this pathology, and a positive correlation between maternal triglycerides levels and neonatal body weight or fat mass has been found in GDM [7,8].

Even when hypercholesterolemia, described in GDM, is not related with the fetal macrosomia, could be related with fetal endothelial dysfunction and later development of cardiovascular diseases in the adulthood [6].

Although lipid traffic through the placenta is restrictive, a correlation between maternal and fetal blood cholesterol in the first and second trimesters of pregnancy has been established, suggesting that maternal cholesterol level could alter normal development of the fetus [9]. In fact it has been reported that due to altered lipid metabolism in the placenta as a result of high maternal blood cholesterol, atherogenesis, a clinical complication commonly appearing in adults, probably begins in fetal life with similar factors altered at the mother, the fetus and the placenta [9, 10].

In this regard, GDM correlates with placental macro and microvascular endothelial dysfunction, also considered as early marker of atherosclerosis, and neonates from GDM pregnancies have significant increase in the aortic intima-media thickness and higher lipid content, both considered as subclinical markers of atherosclerosis, conditions that will potentially increase the atherosclerotic process later in life [11,12].

Since the lack of information in the literature, nothing is yet available about the potential effect of hypercholesterolemia in GDM pregnancies regarding development of endothelial dysfunction and atherosclerosis in human fetoplacental vasculature [6], however cumulative evidence shows that high levels of blood cholesterol modify the endothelial function in different vascular beds, mostly associated with reduced vascular nitric oxide (NO) bioavailability (i.e. the L-arginine/NO pathway) and elevated oxidative stress leading to reduced vascular reactivity, and then vascular reactivity in children and adults [13].

Several changes caused by hypercholesterolemia could explain these alterations including post-transcriptional down-regulation of cationic amino acid transporters (hCATs)-mediated L-arginine transport [14], reduced NO synthase (NOS) expression [15], reduced expression of tetrahydrobiopterin (BH<sub>4</sub>) an NOS cofactor [16], and increased expression and activity of arginases (enzymes that compete by L-arginine with NOS) [17] among others factors that finally leads to reduction of NO synthesis and endothelial dysfunction. Interestingly, these mechanisms have not been evaluated in GDM coursing with hypercholesterolemia [6].

## 2. Hypercholesterolemia in pregnancy

Several reports show that pregnancy is a physiological condition characterized by a progressive, weeks of gestation-dependent increase (reaching 40-50%) in the maternal blood level of cholesterol [1,2]. This phenomenon is known as maternal physiological hypercholesterolemia in pregnancy (MPH), and is considered to be an adaptive response of the mother to satisfy the high cholesterol demand by the growing fetus [3,4].

In the lack of a consensus and currently available information for general population, a mean value calculated from the reported data in the literature rising to ~247 mg/dl of blood cholesterol could represents a state of MPH (see table 1). When a maternal misadaptation to the

cholesterol demand by the fetus occurs, a group of these women develop a pathological condition described as maternal supraphysiological hypercholesterolemia (MSPH) in pregnancy [5]. Unfortunately, the establishment of a cut-point value for this condition is difficult to define because the scarce information in the literature regarding this condition. However, a review of the available information allows establish a MSPH condition when the maternal blood cholesterol at term of pregnancy level is over the 90<sup>th</sup> percentile or establishing a cut-point defined by different authors and based in their findings (Table 1).

With this global lack of information, the prevalence of MSPH in the pregnant population is unknown and could certainly be a consequence of the fact that maternal blood cholesterol level is not routinely evaluated during pregnancy. However, has been reported that the global prevalence for high blood cholesterol level (>200 mg/dl) in non-pregnant women is 40% with a range between 23% (Asia) and 53% (Europe) [18]. Based on this official information from WHO and assuming that pregnancy results in an increase of 40-50% in blood cholesterol [4], it is conceivable that a significant number of women that get pregnant will develop MSPH and who will potentially present an adverse intrauterine condition that could result in facilitating the developing of vascular alterations and atherosclerosis in the growing fetus.

### **2.1. Cholesterol traffic in pregnancy**

Although lipids traffic through the placenta is restrictive and children born from MSPH generally have normal blood cholesterol level [19], a correlation between maternal and fetal blood cholesterol in the first and second trimesters of pregnancy has been established [9,20].

The sources of cholesterol for fetal metabolism along with endogenous production by fetal tissues include transplacental mother-to-fetus transport of cholesterol [9,19,21-26].

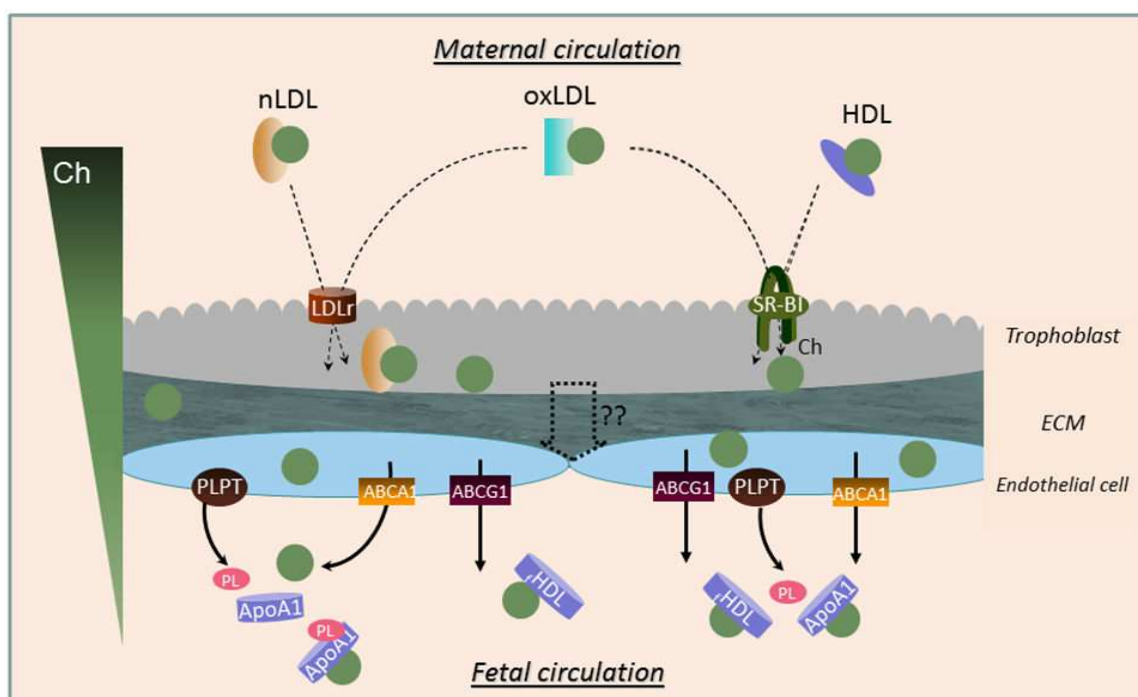
The maternal cholesterol must cross two layers of cells to enter in the fetal circulation, the first one are the trophoblast cells and the second one are the endothelial cells [19,27] (Figure 1).

In the maternal circulation the cholesterol is mainly transported in low density (LDL) and high density (HDL) lipoproteins which interacts with their membrane receptors, the LDL receptor (for native LDL (nLDL) and oxidized LDL (ox LDL)), the lectin-like oxidized low-density lipoprotein receptor-1 (LOX-1, for oxLDL), and scavenger receptor class B type I (SR-BI, for HDL and oxLDL) to deliver the cholesterol content into the cell [28,29]. These lipoprotein receptors are expressed in placental cells including trophoblast and endothelial cells [23,30]. Once in the trophoblast cells, the cholesterol may exit cells secreted as lipoprotein or effluxed from the cellular membrane to extracellular acceptors precursors of mature lipoproteins (i.e., apolipoproteins or discoidal phospholipids) [19]. In the next step, this cholesterol is uptake by endothelial cells to be deliver in the fetal circulation, phenomenon where the expression of cholesterol transporters type ATP binding cassette transporter sub-family A member 1 (ABCA1) and sub-family G member 1 (ABCG1) is determinant since these transporters participate in the efflux of cholesterol to nascent fetal lipoproteins [26,31]. In this scenery the phospholipid transporter protein (PLTP) also participate in the formation of fetal HDL (fHDL) contributing with the efflux of phospholipids to nascent fHDL [26] (Figure 1).

Studied population (n)	Blood cholesterol (mg/dl)		Ref.
	MPH	Cut-point for MSPH	
USA (29)	251	318	[5]
USA (142)	260	300	[157]
USA (553)	250	300	[158]
Canada (59)	248	290	[24]
Mexico (130)	189	-	[159]
Argentina (101)	244	-	[160]
Chile (86)	263	280	[unpublished]
UK (8)	289	-	[161]
UK (40)	315	-	[162]
UK (114)	273	-	[163]
UK (118)	246	-	[163]
UK (178)	264	-	[164]
Italy (82)	-	281	[9]
Italy (156)	-	280	[21]
Italy (72)	205	280	[165]
Italy (22)	286	-	[1]
Norway (12.573)	211	-	[166]
France (73)	242	-	[167]
Germany (150)	253	-	[168]
Spain (45)	225	-	[143]
Spain (66)	259	-	[169]
Portugal (67)	285	-	[170]
Finland (22)	274	-	[171]
Japan (19)	280	-	[172]
China (20)	184	-	[173]
Pakistan (45)	209	-	[174]
Nigeria (222)	204	-	[175]
Tunisia (30)	222	-	[176]
<b>Mean</b>	247		

The values correspond to the third trimester of pregnancy. MPH: maternal physiological and hypercholesterolemia, MSPH: maternal supraphysiological hypercholesterolemia.

**Table 1.** Maternal total cholesterol in MPH and MSPH pregnancies.



**Figure 1.** Mother-to-fetus transport of maternal cholesterol. In the maternal circulation (higher levels of plasma cholesterol) the cholesterol (Ch) is mainly transported in low density lipoproteins (LDL) (natives, nLDL and oxidized, oxLDL) and high density lipoproteins (HDL) lipoproteins and delivered into the trophoblast by LDL receptor (LDL-r) and scavenger receptor class B type I (SR-BI). The cholesterol deliver to extracellular matrix (ECM) and is uptake by endothelial cells through unknown mechanisms to finally, be delivered to the fetal circulation by the of cholesterol transporters type ATP binding cassette transporter sub-family A member 1 (ABCA1) and sub-family G member 1 (ABCG1), which together with phospholipid (PL)-transfer protein (PLPT) contribute with the assembly of fetal lipoproteins. In fetal circulation (lower levels of plasma cholesterol) cholesterol is deliver to acceptors as ApoA1 and nascent fetal HDL (fHDL).

Thus the mother-*to*-fetus transport of cholesterol seems to be a controlled process that is crucial in fetal development; however the effect of a supraphysiological level of maternal cholesterol will modify the traffic of cholesterol increasing the risk of developing fetal vascular anomalies such as those seen in atherosclerosis [31].

## 2.2. Consequences of MSPH in the fetus

Studies in aortas from spontaneously aborted human fetuses and premature newborns (24-30 weeks of gestation) demonstrate that offspring from mothers with MSPH in pregnancy exhibit more and larger aortic lesion which were positive for almost one marker of atherosclerosis among the presence of macrophages and foam cell, LDL, oxLDL and oxidation-specific epitopes [9]. These data were additionally supported by another autopsy study that determined that children (1-13 years old) of mothers with MSPH in pregnancy exhibit faster progression of atherosclerotic lesions [21].

At present, the effect of MSPH have been evaluated as atherosclerosis in fetal arteries but the vascular effects of MSPH could be determined in placental vessels since its cells are indirectly exposed to maternal cholesterol (see section *Cholesterol traffic in pregnancy*). Interestingly, it has

been shown that MSPH is associated with increased expression of placental genes related to cholesterol metabolism (i.e. fatty acid synthase (FAS), sterol regulatory element-binding protein 2 (SREBP2)), thus exposing the fetus to an altered lipid environment and eventually promoting vascular alterations [24]. Additionally, increased level of maternal cholesterol and LDL leads to down-regulation of LDL receptor expression in whole placenta homogenized without changes in the expression of HDL receptor (SR-BI) [32], suggesting that the increase in the LDL concentration in the maternal blood induce the regulation of the LDL receptor expression. Interestingly these alterations are not related with changes in the newborn lipid levels, in fact normal levels of LDL and total cholesterol are determined at birth in the fetal blood of newborns from mothers with MSPH.

These data provide evidence for the potential effect of MSPH on the placenta and its consequences for the fetus where vascular lesion progression is triggered. However, even knowing this available information nothing is reported regarding whether abnormal maternal blood cholesterol level leads to placental vascular dysfunction [10,33].

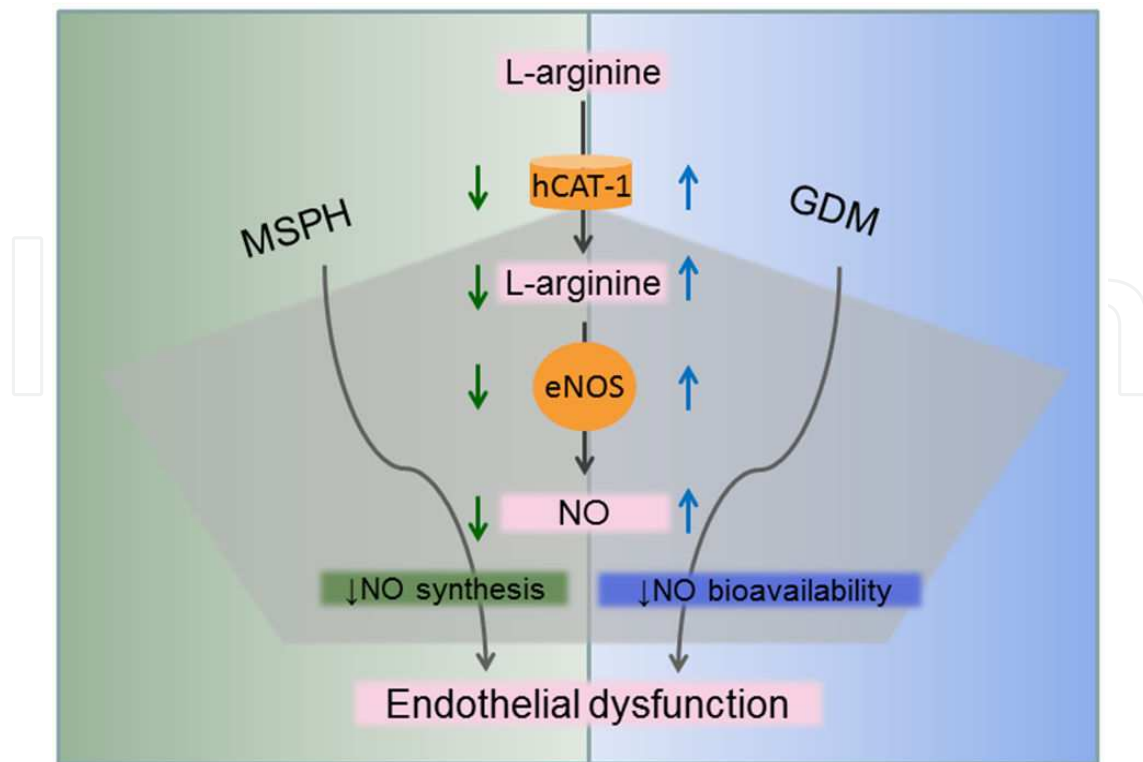
### 3. Endothelial function in normal pregnancies

The placenta is a physical and metabolic barrier between the fetal and maternal circulation. The normal development and function of the placenta and the umbilical cord are crucial to sustain the adequate fetal development and growth [34]. The human fetoplacental circulation under physiological conditions exhibits a high blood flow and low vascular resistance [35]. Since it lacks of autonomic innervation [36] the equilibrium between the synthesis, release and bioavailability of vasoconstrictors and vasodilators circulating and locally released, such as NO and adenosine, are crucial to maintain the control of fetoplacental hemodynamics [37,38]. In a physiological context, different pathologies of pregnancy such as GDM [38,39], intrauterine growth restriction (IUGR) [40] or preeclampsia [41], exhibits altered synthesis and/or bioavailability of NO leading to changes in blood flow of the human placenta thus limiting fetal growth and development [37,38,42]. These conditions produce an imbalance or loss of essential endothelial functions leading to altered blood flow in the fetoplacental unit mainly associated with altered NO synthesis and membrane transport of the semi-essential cationic amino acid L-arginine, i.e., the 'endothelial L-arginine/NO pathway' (Figure 2) [35,42,43].

#### 3.1. Endothelial L-arginine/NO signaling pathway

Synthesis of NO requires active NOS, a group of enzymes conformed by, at least, three isoforms, i.e., neuronal NOS (nNOS or type 1), inducible NOS (iNOS or type 2) and endothelial NOS (eNOS or type 3), of which mainly eNOS is expressed in endothelial cells [43,44]. The NO diffuses from endothelium to vascular smooth muscle cells leading to cyclic GMP (cGMP)-dependent vasodilatation [45].

Activity of NOS may depend on the ability of endothelial cells to take up its specific substrate L-arginine via a variety of membrane transport systems [42,43,46,47]. L-Arginine is taken up into the endothelial cells through the membrane transport systems  $y^+$  (cationic amino acid



**Figure 2.** Endothelial L-Arginine/NO pathway regulation by GDM and MSPH. In human endothelial cells L-arginine is mainly taken up via cationic amino acid transporter 1 (hCAT-1). Then, L-arginine is metabolized in nitric oxide (NO) and L-citrulline by endothelial NO synthase (eNOS). In GDM is described an increase (blue up arrows) in the elements in L-Arginine/NO pathway, but with a decrease in NO bioavailability leading endothelial dysfunction. As seen in other vascular beds, MSPH leads to decreased (green down arrows) hCAT-1 and eNOS expression; and NO synthesis leading also to endothelial dysfunction.

transporters family, CATs),  $y^+L$  (very high affinity transporters),  $b^{0,+}$  and  $B^{0,+}$  ( $Na^+$ -independent and dependent, respectively) [48,49,50]. CATs is a family of membrane transporters with at least 5 isoforms identified in human tissues, i.e., human CAT-1 (hCAT-1), hCAT-2A, hCAT-2B, hCAT-3 and hCAT-4 [43,51]. In endothelial cells from the human placenta such as human umbilical vein endothelial cells (HUVEC) and human placental microvascular endothelial cells (hPMEC), only hCAT-1 and hCAT-2B isoforms like transport have been identified, the first exhibiting low-capacity and high-affinity, and the second exhibiting higher-capacity and lower-affinity [41,51]. Moreover, eNOS activity seems to depend on the ability of these cells to take up L-arginine mainly via hCAT-1 and/or hCAT-2B [40, 33, 52]. Interestingly in pathological conditions such as GDM the L-arginine/NO pathway is highly up-regulated in HUVEC [6, 39, 53, 54] (Figure 2).

#### 4. Hypercholesterolemia and endothelial L-arginine/NO pathway

Endothelial dysfunction and reduced NO seems are considered early markers in the development of cardiovascular disease [55-58]. Thus, studies designed to evaluate the impact of



hypercholesterolemia (in non-placental vessels) have determined that this pathological condition induces endothelial dysfunction in vessels of the macro and microcirculation, but the biological effects may differ between both vascular beds [59-60]. It has been shown that high levels of total cholesterol and oxLDL impair endothelial function increasing the production of the vasoconstrictor endothelin-1 [61-62] and reducing NO bioavailability [13,63-67], alterations that have been associated with impaired endothelium-dependent relaxation [68-73]. Therefore, alterations in cholesterol levels leading to endothelial dysfunction in different vascular beds have been associated with molecular changes in the expression and activity of different component of the L-arginine/NO pathway, thus decreasing the production or bioavailability of NO (Table 2). However, no studies have addressed whether elevated maternal blood cholesterol modulate L-arginine/NO pathway and endothelial function in placental endothelial cells from pregnancies coursing with MSPH or pregnancy diseases associated with increased levels of cholesterol as GDM or preeclampsia [12,35].

#### **4.1. eNOS expression and activity in hypercholesterolemia**

Hypercholesterolemia is associated with decreased expression of eNOS in aortic rings of hypercholesterolemic rabbits [58] and in human saphenous vein endothelial cell, porcine aortic endothelial cells and HUVEC expose to high concentration of nLDL or ox-LDL [15,75,76], an effect that is reversed by restitution of normal blood cholesterol level (e.g., with the use of statins). The mechanism behind this effect of hypercholesterolemia on eNOS expression is not well understood and few studies have proposed a time- and concentration-dependent decrease in eNOS mRNA level involving transcriptional inhibition and reduced mRNA stability (i.e., reducing eNOS mRNA half-life) [15,75,76].

Additionally to down regulation of eNOS expression, high levels of cholesterol are also associated with changes in eNOS cellular localization and function, a phenomenon related with up-regulation of the protein caveolin [77-83]. In the endothelial cell eNOS targets to caveolae [70,71] where it is functionally inhibited by binding to caveolin [84-87]. Optimal eNOS activity occurs when the eNOS-caveolin complex interaction is disrupted by calcium-calmodulin binding to eNOS-caveolin [87]. It has been shown that caveolin expression is regulated by cholesterol increasing eNOS-caveolin complex formation, and diminishing NO production [88-90].

#### **4.2. Asymmetrical dimethylarginine (ADMA) availability in hypercholesterolemia**

ADMA, an arginine metabolite proposed as endogenous inhibitor of eNOS [91-95], is increased in hypercholesterolemic monkeys [92] and in human endothelial cells incubated with high concentration of nLDL and oxLDL [96]. The mechanisms involved in this phenomenon are the up-regulation of the expression of protein arginine N-methyl transferases (PRMTs), which are involved in the synthesis of ADMA and decreased activity of dimethylarginine dimethylaminohydrolase (DDAH), an enzyme responsible of ADMA degradation [78,82,83]. Moreover, the regulation of ADMA is relevant in the atherogenic process and extensive data have shown a good correlation between plasmatic levels of ADMA and the presence of atherosclerosis [92].

Element	Gestational Diabetes Mellitus			Non-pregnancy		
	Cell type	Effect	References	Cell type	Effect	References
hCATs expression	HUVEC	Increased	[43]	EAhy926	Increased	[113]
				rAR	Increased	[114]
hCATs activity	HUVEC	Increased	[43]	EAhy926	Increased	[113]
				rAR	Reduced	[14]
				bAEC	Reduced	[111]
				pAEC	Reduced	[74]
				HUVEC	Unaltered	[109]
				HUVEC	Unaltered	[110]
eNOS expression	HUVEC	Increased	[28,43]	hSVEC	Reduced	[15]
				hPT	Increased	[177]
	hSVEC	Reduced	[76]	rbAS	Increased	[75]
				rbAS	Reduced	[152]
				pAEC	Reduced	[180]
eNOS activity	HUVEC	Increased	[28,29,43]	hSVEC	Reduced	[15]
				hVT	Unaltered	[178]
	hSVEC	Reduced	[76]	rbAS	Reduced	[152]
				rbAS	Reduced	[76]
				pAEC	Reduced	[74]
rAC	Reduced	[181]				
NO level	HUVEC	Increased	[179]	hSVEC	Reduced	[15]
Arginase I				hPBMC	Increased	[182]
Arginase II				hAEC	Increased	[17,114,115]
				mAEC	Increased	[114,115]

hCATs, human cationic amino acid transporters; eNOS, endothelial nitric oxide synthase; NO, nitric oxide; HUVEC, human umbilical vein endothelial cell; hPT, human placental tissue; hVT, human villous tissue; EAhy 926, human endothelial cell line EAhy 926; rAR, rat aortic ring; bAEC, bovine aortic endothelial cell; pAEC, porcine aortic endothelial cell; hSVEC, human saphenous vein endothelial cell; rbAS, rabbit aortic segment; rAC, rat aortic cell; hPBMC, peripheral blood mononuclear cells; hAEC, human aortic endothelial cell; mAEC, mouse aortic endothelial cell. Table modified from reference 6.

**Table 2.** Effect of GDM and hypercholesterolemia on endothelial L-Arginine/NO pathway.

Thus, this is a different way by which increased levels of cholesterol leads to a reduction in NO synthesis.

### 4.3. Tetrahydrobiopterin (BH<sub>4</sub>) availability in hypercholesterolemia

A reduced expression of the eNOS cofactor BH<sub>4</sub> leads to deficient activation (or even uncoupling) of eNOS, a phenomenon characterized by eNOS-reduction of molecular oxygen by a no longer coupled L-arginine oxidative mechanism resulting in generation of superoxide anion rather than NO [98]. This phenomenon contributes to vascular oxidative stress and endothelial damage and dysfunction [16]. Hypercholesterolemic mice and rabbit exhibit reduced level of BH<sub>4</sub> in the aorta and myocardium [99,100], a phenomenon related with endothelial dysfunction and major progression of atherosclerosis. Additionally, it has been demonstrated that BH<sub>4</sub> supplementation improves the endothelial function in hypercholesterolemic patients [101,102], suggesting that this cofactor is reduced in this pathological condition. Endothelial cells from the human placenta vasculature express functional BH<sub>4</sub> which is reduced with the progress of pregnancy by a mechanism involving lower activity of guanosine triphosphate cyclohydrolase I (GTPCH) and 6-pyruvoyl tetrahydropterin synthase (PTPS), key enzymes involved in BH<sub>4</sub> synthesis [103,104]. Alternatively, in other cell types, a reduced level of BH<sub>4</sub> dependent of down-regulation of GTPCH expression has been associated with hypercholesterolemia in rat macrophages and smooth muscle cells [105,106].

### 4.4. L-Arginine transport in hypercholesterolemia

Decreased bioavailability of L-arginine could result from reduced expression and/or altered cellular localization of hCATs, as reported for hCAT-1 and potentially hCAT-2B in HUVEC [53,54,107,108]. Interestingly, it was initially shown that hCAT-1-like transport was unaltered by oxLDL in HUVEC cultures [109,110]. However, no kinetic parameters were addressed in these studies opening the possibility that L-arginine transport at a unique fixed concentration of this amino acid (100 μM) [109] could be insensible to oxLDL, or that a long period of incubation for L-arginine uptake (1-24 hours) [110] will not be a condition close to initial velocity for transport, something required for this type of analysis [49,51]. Additional studies in other types of endothelial cells show that LDL (native or oxLDL) reduces L-arginine transport in aortic endothelium from hypercholesterolemic rats, involving protein kinase C [14]; and bovine aortic endothelium where a maximal transport capacity ( $V_{max}/K_m$ ) [49] is reduced [111,112]. Interestingly, human aortic endothelial cells exposed to nLDL/oxLDL exhibit decreased intracellular content of L-arginine, a phenomenon explained as resulting from post-translational down-regulation of CAT1 and increased CAT1 internalization [102]. In addition, and highlighting the involvement of L-arginine transport in placental vascular reactivity, recent studies suggest that L-arginine transport mediated by hCAT-1 will be a mechanism limiting human placental vascular reactivity since reduced transport (by the use of *N*-ethylmaleimide) or cross-inhibition (by L-lysine) of hCATs leads to reduced insulin-induced dilatation of human umbilical vein rings from normal pregnancies [54].

### 4.5. Arginases expression in hypercholesterolemia

Up-regulation of arginases (isoforms I and II) is another mechanism by which NO synthesis is proposed to be reduced leading to placental endothelial dysfunction. Arginases are enzyme competing by L-arginine with eNOS [17,114,115], favoring conversion of L-

arginine into L-ornithine and urea. Therefore, an increase in arginases activity will limit the availability of L-arginine to be metabolized by eNOS for NO synthesis. Interestingly, a link between hypercholesterolemia and arginase I and II expression has been demonstrated in mice [115] and in human aortic endothelial cells [116] where oxLDL induces an overexpression of arginases and a reduction of total eNOS protein abundance associated with lower NO production [114], mostly by the interaction with LOX-1 receptor and the activation of the small GTPase RhoA and Rho A kinase (ROCK) signaling pathway [17]. Interestingly, the reduction of arginases activity caused by statins in hypercholesterolemic subjects improves the endothelial function [117]. These findings show that arginases could play a role in the modulation of endothelial function, most likely regarding NO synthesis by competing for L-arginine with eNOS.

## 5. Gestational diabetes mellitus

GDM is a syndrome characterized by glucose intolerance with onset or first recognition during pregnancy [118-120]. Clinical manifestations of GDM have been attributed mainly to the condition of hyperglycemia, hyperlipidemia, hyperinsulinemia, and fetoplacental endothelial dysfunction [34,37,119,121,123]. GDM is also associated with abnormal fetal development and perinatal complications, such as macrosomia, neonatal hypoglycemia, and neurological disorders [121]. This syndrome occurs with a high incidence, depending on diagnostic criteria used, ranging between 5 and 15% of pregnant women in developing [124,125] and developed countries [120,126-128].

Altered vascular reactivity is a characteristic of GDM and is due to endothelial dysfunction at the micro and macro fetoplacental vasculature [34,37,129-134].

Even when hyperglycaemia is the principal factor leading to endothelial dysfunction, other factors are involved including hyperinsulinemia and the extracellular nucleoside adenosine level [39,133,134]. Since GDM is associated with MSPH, this factor could also contribute with this phenomenon although the effect is actually unknown.

### 5.1. Endothelial function in GDM and L-arginine/NO pathway

It has been reported that the NO level in the human umbilical vein blood is increased in GDM [127] and that in HUVEC from GDM the synthesis of NO is increased [39,53, 135, 136]. These findings were associated with a constitutive increase in the number of copies for eNOS mRNA, as well as increased eNOS protein level and activity. Other studies show that in HUVEC isolated from GDM the L-arginine transport is increased due to higher maximal velocity ( $V_{max}$ ) for transport, most likely resulting from increased expression of hCAT-1 [53,133]. Even when the synthesis of NO is increased in GDM cells, the bioavailability of this vasodilator is reduced leading to an state of endothelial dysfunction [6,34,39, 123] (Figure 2). Thus, the vascular reactivity of umbilical vein rings from GDM is lower compare with rings from normal pregnancies [39]. This phenomenon has been suggested to result from a less reactive umbilical vein due to a tonic and basal increased state of

vasodilation by over-release and/or accumulation of adenosine, a nucleoside that induce vascular relaxation, in the umbilical vein blood [39].

## 6. Dyslipidemia in GDM

GDM is a pathological condition also characterized by maternal dyslipidemia, alterations that affect directly the fetal development and growth [123].

Dyslipidemia is defined as elevated levels of triglycerides (hypertriglyceridemia) and total blood cholesterol (hypercholesterolemia) including increased LDL and reduced HDL cholesterol [137]. Dyslipidemia is recognized as the main risk factor for development of CVD [137,139]. Additionally, GDM has also been established as a significant risk factor to fetal programming of metabolic syndrome [140-142] and thus predisposing to accelerate the development of CVD in the adult life [141-146].

Interestingly, most of pregnancies that develop GDM course with dyslipidemia [7,24,147] (Table 3) and thus could be feasible to found a pathologic link between dyslipidemia in pregnancies with GDM and the development of CVD later in life.

GDM is related with fetal macrosomia and endothelial dysfunction and interestingly both characteristic could be related with the associated dyslipidemia. The association between dyslipidemia and macrosomia regards hypertriglyceridemia more than hypercholesterolemia; in fact, a positive correlation between maternal triglycerides and neonatal body weight or fat mass has been found in GDM [7,141,142]. In the other hand, hypercholesterolemia could contribute with the endothelial dysfunction described in the pathology [6,142,149]. Thus GDM could play a role in the fetal programming of adult CVD not only by the classical alterations mainly triggered by hyperinsulinemia, hyperglycaemia and changes in nucleoside extracellular concentration, but also by hypercholesterolemia associated with this pathology [6,142,149]. However, no studies have addressed whether elevated maternal blood cholesterol in GDM modulate endothelial function in placental endothelial cells [33].

### 6.1. Cholesterol metabolism in GDM

The increased levels of maternal cholesterol in GDM (Table 3) are related with alterations in the expression of proteins involved in lipid and cholesterol homeostasis [24,150,151].

Although MSPH is associated with decreased expression of LDL receptor in the placenta, the effect of GDM-associated dyslipidemia on lipoprotein receptors expression is unknown [24,32]. A study of microarray profile determined changes in the expression of multiple genes involved in lipid and cholesterol metabolism in placental tissue of pregnancies coursing with GDM. These genes include the fatty acid coenzyme A ligase, long chain 2, 3 and 4 (FACL2,3,4) that catalyze the conversion of fatty acids into fatty acyl-CoA esters (precursors for the synthesis of triglycerides, cholesterol, and membrane phospholipids), additionally 3-hydroxy-3-methylglutaryl-Coenzyme A reductase (HMGCR), 3-hydroxy-3-methylglutaryl-

	1 <sup>o</sup> trimester	2 <sup>o</sup> trimester	3 <sup>o</sup> trimester	Reference
TG	101*	142*	187*	[147]
(mg/dl)		286*	271*	[183]
			226*	[184]
			260*	[185]
			268	[24]
			220	[158]
			203	[186]
		214		[187]
Ch	203*	226*	281*	[147]
(mg/dl)		241	224	[183]
			242	[184]
			267	[24]
			197	[158]
			243	[185]
		246		[187]
LDL		145	130	[183]
(mg/dl)		129		[187]
			130	[184]
			148 <sup>†</sup>	[185]
			143	[24]
			197	[158]

Values of cholesterol (Ch) and triglycerides (TG) in GDM pregnancies were compared with those of normal pregnancy. \*: level increased compared with a control group without GDM. †: level decreases compared with a control group without GDM. LDL: low-density lipoprotein.

**Table 3.** Maternal lipids levels GDM pregnancies.

Coenzyme A synthase (HMGCS 1) among other genes involved in the novo synthesis of cholesterol were also regulated [150] and even when in this study the level of cholesterol were not determined among normal and GDM pregnancies, these data suggest that GDM leads to changes in genes related with cholesterol metabolism in the placenta. Previously was described that MSPH associates with increased expression of FAS and SREBP2 in the placenta, while the effect in FAS was observed also in placental cells from GDM without changes in SREBP2 expression [24].

These data suggest that both MSPH and GDM associates with changes in key element in the lipids metabolism, however, if MSPH potentiate the effect of GDM over these parameters is unknown [6].

Another lipid modulator modified by GDM in placental cells is PLTP, a key protein involved in the metabolism of fetal HDL. PLTP is expressed in endothelial cells of the placental vasculature and is regulated as ABCA1 and ABCG1 by liver X receptor (LXR) nuclear receptors [26,152,153]. Interestingly diabetes leads to increased levels of the principal ligand of LXR, the oxysterols [154] and GDM associates with up-regulation of PLTP in endothelial cells of the placenta [151] mainly due to the hyperinsulinemia and hyperglycaemia related with GDM. The increased expression of PLTP could be a key phenomenon associated with the increased concentration of HDL described in newborns from GDM [11,151]. Additionally, the increased expression of PLTP in placental endothelial cells could affect the maternal to fetal cholesterol transport, a phenomenon not yet evaluated and potential worsen by conditions as MSPH where the mother-to-fetus cholesterol transport may be altered almost in the first months of pregnancies when the levels maternal cholesterol correlates with the fetal ones [9].

## 6.2. Hypercholesterolemia in GDM and endothelial dysfunction

As was previously discussed, physiological increase in the levels of maternal cholesterol is considered to be an adaptive response of the mother to satisfy the high lipids demand by the growing fetus. The misadaptation to this condition leads to develop MSPH a phenomenon associated with the earlier development of fetal atherosclerosis and with reduced endothelial function of the umbilical vein [6].

Additionally and regarding with the development of atherosclerosis, it is recognized that GDM correlates with endothelial dysfunction [34,39] and neonates of pregnancies coursing with GDM have significant increase in aortic and umbilical artery intima-media thickness (IMT) and higher lipid content, both markers of subclinical atherosclerosis that could increase the atherosclerotic process later in life [12,155,156].

The effect of GDM in the aortic IMT of newborns was assayed and an increased intimal-medial ratio was determined. Interestingly the IMT was evaluated in newborns of pregnancies coursing with GDM and increased levels of total cholesterol and LDL compared with the control group [12]. Thereby may be possible to found a potential effect of MSPH in this phenomenon. Similar findings were found in fetus in the lasts weeks of gestation where the IMT was evaluated in umbilical artery, where umbilical IMT was increased in arteries from GDM pregnancies, however the potential effect of maternal cholesterol was not evaluated [156].

Unfortunately, nothing is yet available regarding the potential effects of MSPH in pregnancies coursing with GDM on the development of endothelial dysfunction and atherosclerosis in placental and eventually in fetal vessels at birth, a phenomenon that could lead to a potentiation of GDM-associated fetoplacental endothelial dysfunction.

## 7. Concluding remarks

MSPH is a risk factor promoting the development of atherosclerosis in the growing fetus and in the children, however the effects of this condition in fetoplacental endothelium is unknown even when increased levels of maternal cholesterol could lead to alterations in the hCAT-mediated L-arginine transport and eNOS-synthesis of NO (i.e., the endothelial L-arginine/NO signaling pathway) such as occurs in other vascular beds exposed to high cholesterol levels.

GDM is a condition that course with alterations of the L-arginine/NO signaling pathway in the human fetoplacental vasculature, phenomenon resulting in abnormal bioavailability of NO leading to altered vascular reactivity and changes in umbilical vessels blood flow with consequences in the fetal growth and development.

Interestingly, some pregnancies coursing with GDM associates with MSPH and the possibility that the observed fetoplacental endothelial dysfunction results from a potentiation of the classical factor associated with GDM and the increased levels of cholesterol is likely.

Further studies are required to elucidate whether pregnancies coursing with GDM and MSPH have different effect in placental endothelial function compare with those coursing with GDM and normal levels of maternal cholesterol because it could be possible find a different mechanism involved in both cases.

This may contribute to understand the mechanisms related with the vascular dysfunction associated with GDM and allow establishing a better knowledge based- management of the mother and the newborn.

## Acknowledgements

We are thankful to the personnel at the Hospital Clínico Pontificia Universidad Católica de Chile labour ward for their support in the supply of placentas. This research was funded by Fondo Nacional de Desarrollo Científico y Tecnológico (FONDECYT 1110977, 11110059, 3130583), Programa de Investigación Interdisciplinario (PIA) from Comisión Nacional de Investigación en Ciencia y Tecnología (CONICYT, Anillos ACT-73) (Chile) and CONICYT Ayuda de Tesis (CONICYT AT-24120944). EG-G holds a CONICYT-PhD (Chile) fellowship. FP was the recipient of a postdoctoral position (CONICYT PIA Anillos ACT-73 postdoctoral research associate at CMPL, Pontificia Universidad Católica de Chile (PUC)). CDM was the recipient of an undergraduate research position (CONICYT PIA Anillos ACT-73 undergraduate researcher at CMPL, PUC).



## Author details

A. Leiva\*, C Diez de Medina, E. Guzmán-Gutierrez, F. Pardo and L. Sobrevia

\*Address all correspondence to: aaleiva@puc.cl, sobrevia@med.puc.cl

Cellular and Molecular Physiology Laboratory (CMPL), Division of Obstetrics and Gynecology, Faculty of Medicine, School of Medicine, Pontificia Universidad Católica de Chile, Chile

## References

- [1] Brizzi P, Tonolo G, Esposito F, Puddu L, Dessole S, Maioli M, et al. (1999) Lipoprotein metabolism during normal pregnancy. *American journal of obstetrics and gynecology* 181: 430-434.
- [2] Avis HJ, Hutten BA, Twickler MT, Kastelein JJ, van der Post JA, Stalenhoef AF, et al. (2009) Pregnancy in women suffering from familial hypercholesterolemia: a harmful period for both mother and newborn? *Current opinion in lipidology* 20:484-490.
- [3] Schaefer-Graf UM, Meitzner K, Ortega-Senovilla H, Graf K, Vetter K, Abou-Dakn M, et al. (2011) Differences in the implications of maternal lipids on fetal metabolism and growth between gestational diabetes mellitus and control pregnancies. *Diabetic medicine* 28:1053-1059.
- [4] Basaran A (2009) Pregnancy-induced hyperlipoproteinemia: review of the literature. *Reproductive sciences* 16:431-437.
- [5] Montes A, Walden CE, Knopp RH, Cheung M, Chapman MB, Albers JJ (1984) Physiologic and supraphysiologic increases in lipoprotein lipids and apoproteins in late pregnancy and postpartum. Possible markers for the diagnosis of "prelipemia". *Arteriosclerosis* 4:407-417.
- [6] Leiva A, Pardo F, Ramirez MA, Farias M, Casanello P, Sobrevia L (2011) Fetoplacental vascular endothelial dysfunction as an early phenomenon in the programming of human adult diseases in subjects born from gestational diabetes mellitus or obesity in pregnancy. *Experimental diabetes research* 2011:349286.
- [7] Herrera E, Ortega-Senovilla H (2010) Disturbances in lipid metabolism in diabetic pregnancy - Are these the cause of the problem? *Best practice and research. Clinical endocrinology & metabolism* 24:515-525.
- [8] Son GH, Kwon JY, Kim YH, Park YW (2010) Maternal serum triglycerides as predictive factors for large-for-gestational age newborns in women with gestational diabetes mellitus. *Acta Obstetrics and Gynecology Scandinav* 89:700-704.

- [9] Napoli C, D'Armiento FP, Mancini FP, Postiglione A, Witztum JL, Palumbo G, et al. (1997) Fatty streak formation occurs in human fetal aortas and is greatly enhanced by maternal hypercholesterolemia. Intimal accumulation of low density lipoprotein and its oxidation precede monocyte recruitment into early atherosclerotic lesions. *The Journal of clinical investigation* 100:2680-2690.
- [10] Palinski W, Napoli C (2002) The fetal origins of atherosclerosis: maternal hypercholesterolemia, and cholesterol-lowering or antioxidant treatment during pregnancy influence in utero programming and postnatal susceptibility to atherogenesis. *FASEB journal* 16:1348-1360.
- [11] Merzouk H, Madani S, Prost J, Loukidi B, Meghelli-Bouchenak M, Belleville J (1999) Changes in serum lipid and lipoprotein concentrations and compositions at birth and after 1 month of life in macrosomic infants of insulin-dependent diabetic mothers. *European Journal of Pediatrics* 158:750-756.
- [12] Koklu E, Akcakus M, Kurtoglu S, Koklu S, Yikilmaz A, Coskun A, et al. (2007) Aortic intima-media thickness and lipid profile in macrosomic newborns. *European Journal of Pediatrics* 166:333-338.
- [13] Landmesser U, Hornig B, Drexler H (2000) Endothelial dysfunction in hypercholesterolemia: mechanisms, pathophysiological importance, and therapeutic interventions. *Seminars in Thrombosis and Hemostasis* 26:529-537.
- [14] Schwartz IF, Ingbir M, Chernichovski T, Reshef R, Chernin G, Litvak A, et al. (2007) Arginine uptake is attenuated, through post-translational regulation of cationic amino acid transporter-1, in hyperlipidemic rats. *Atherosclerosis* 194:357-363.
- [15] Liao JK, Shin WS, Lee WY, Clark SL (1995) Oxidized low-density lipoprotein decreases the expression of endothelial nitric oxide synthase. *The Journal of Biological Chemistry* 270:319-324.
- [16] Alp NJ, Channon KM (2004) Regulation of endothelial nitric oxide synthase by tetrahydrobiopterin in vascular disease. *Arteriosclerosis, Thrombosis, and Vascular Biology* 24:413-420.
- [17] Ryoo S, Lemmon CA, Soucy KG, Gupta G, White AR, Nyhan D, et al. (2006) Oxidized low-density lipoprotein-dependent endothelial arginase II activation contributes to impaired nitric oxide signaling. *Circulation research* 99:951-960.
- [18] World Health Organization (2010) Global status report on non-communicable diseases 1:9-31
- [19] Woollett LA (2011) Review: Transport of maternal cholesterol to the fetal circulation. *Placenta* 32:S218-S221.
- [20] Badruddin SH, Lalani R, Khurshid M, Molla A, Qureshi R, Khan MA (1990) Serum cholesterol in neonates and their mothers. A pilot study. *Journal of Pakistan Medical Association* 40:108-109.

- [21] Napoli C, Glass CK, Witztum JL, Deutsch R, D'Armiento FP, Palinski W (1999) Influence of maternal hypercholesterolaemia during pregnancy on progression of early atherosclerotic lesions in childhood: Fate of Early Lesions in Children (FELIC) study. *Lancet* 354:1234-1241.
- [22] Battaile KP, Steiner RD (2000) Smith-Lemli-Opitz syndrome: the first malformation syndrome associated with defective cholesterol synthesis. *Molecular genetics and metabolism* 71:154-162.
- [23] Wadsack C, Hammer A, Levak-Frank S, Desoye G, Kozarsky KF, Hirschmugl B, et al. (2003) Selective cholesteryl ester uptake from high density lipoprotein by human first trimester and term villous trophoblast cells. *Placenta* 24:131-143.
- [24] Marseille-Tremblay C, Ethier-Chiasson M, Forest JC, Giguere Y, Masse A, Mounier C, et al. (2008) Impact of maternal circulating cholesterol and gestational diabetes mellitus on lipid metabolism in human term placenta. *Molecular reproduction and development* 75:1054-1062.
- [25] Jenkins KT, Merkens LS, Tubb MR, Myatt L, Davidson WS, Steiner RD, et al. (2008) Enhanced placental cholesterol efflux by fetal HDL in Smith-Lemli-Opitz syndrome. *Molecular genetics and metabolism* 94:240-247.
- [26] Stefulj J, Panzenboeck U, Becker T, Hirschmugl B, Schweinzer C, Lang I, et al. (2009) Human endothelial cells of the placental barrier efficiently deliver cholesterol to the fetal circulation via ABCA1 and ABCG1. *Circulation research* 104:600-608.
- [27] Desoye G, Gauster M, Wadsack C. (2011) Placental transport in pregnancy pathologies. *American Journal of Clinical Nutrition*. 94:S1896-S1902.
- [28] Chen M, Masaki T, Sawamura T (2002) LOX-1, the receptor for oxidized low-density lipoprotein identified from endothelial cells: implications in endothelial dysfunction and atherosclerosis. 95:89-100.
- [29] Mineo C, Deguchi H, Griffin JH, Shaul PW (2006) Endothelial and antithrombotic actions of HDL. *Pharmacology and Therapeutics* 98:1352-1364.
- [30] Marsche G, Levak-Frank S, Quehenberger O, Heller R, Sattler W, Malle E (2001) Identification of the human analog of SR-BI and LOX-1 as receptors for hypochlorite-modified high density lipoprotein on human umbilical venous endothelial cells. *FASEB journal* 15:1095-1097.
- [31] Palinski W, Nicolaidis E, Liguori A, Napoli C (2009) Influence of maternal dysmetabolic conditions during pregnancy on cardiovascular disease. *Journal of cardiovascular translational research* 2:277-285.
- [32] Ethier-Chiasson M, Duchesne A, Forest JC, Giguere Y, Masse A, Mounier C, et al. (2007) Influence of maternal lipid profile on placental protein expression of LDLr and SR-BI. *Biochemical and biophysical research communications* 359:8-14.

- [33] Joles JA (2011) Crossing borders: linking environmental and genetic developmental factors. *Micricirculation* 18:298-303.
- [34] Sobrevia L, Abarzua F, Nien JK, Salomon C, Westermeier F, Puebla C, et al. (2011) Review: Differential placental macrovascular and microvascular endothelial dysfunction in gestational diabetes. *Placenta* 32:S159-S164.
- [35] Myatt L (2010) Review: Reactive oxygen and nitrogen species and functional adaptation of the placenta. *Placenta* 31:S66-S69.
- [36] Marzioni D, Tamagnone L, Capparuccia L, Marchini C, Amici A, Todros T, et al. (2004) Restricted innervation of uterus and placenta during pregnancy: evidence for a role of the repelling signal Semaphorin 3A. *Developmental Dynamics* 231:839-848.
- [37] Westermeier F, Puebla C, Vega JL, Farias M, Escudero C, Casanello P, et al. (2009) Equilibrative nucleoside transporters in fetal endothelial dysfunction in diabetes mellitus and hyperglycaemia. *Current Vascular Pharmacology* 7:435-449.
- [38] Farias M, Puebla C, Westermeier F, Jo MJ, Pastor-Anglada M, Casanello P, et al. (2010) Nitric oxide reduces SLC29A1 promoter activity and adenosine transport involving transcription factor complex hCHOP-C/EBPalpha in human umbilical vein endothelial cells from gestational diabetes. *Cardiovascular Research* 86:45-54.
- [39] Westermeier F, Salomon C, Gonzalez M, Puebla C, Guzman-Gutierrez E, Cifuentes F, et al. (2011) Insulin Restores Gestational Diabetes Mellitus-Reduced Adenosine Transport Involving Differential Expression of Insulin Receptor Isoforms in Human Umbilical Vein Endothelium. *Diabetes* 60:1677-1687.
- [40] Casanello P, Sobrevia L (2002) Intrauterine growth retardation is associated with reduced activity and expression of the cationic amino acid transport systems  $\gamma^+$ /hCAT-1 and  $\gamma^+$ /hCAT-2B and lower activity of nitric oxide synthase in human umbilical vein endothelial cells. *Circulation Research* 91:127-134.
- [41] Escudero C, Sobrevia L (2008) A hypothesis for preeclampsia: adenosine and inducible nitric oxide synthase in human placental microvascular endothelium. *Placenta* 29:469-483.
- [42] Casanello P, Escudero C, Sobrevia L (2007) equilibrative nucleoside (ENTs) and cationic amino acid (CATs) transporters: implications in fetal endothelial dysfunction in human pregnancy diseases. *Current Vascular Pharmacology* 5:69-84.
- [43] Sobrevia L, Gonzalez M (2009) A role for insulin on L-arginine transport in fetal endothelial dysfunction in hyperglycaemia. *Placenta* 7:467-474.
- [44] Alderton WK, Cooper CE, Knowles RG (2001) Nitric oxide synthases: structure, function and inhibition. *Biochemical Journal* 357: 593-615.
- [45] Moncada S, Higgs EA (2006) Nitric oxide and the vascular endothelium. *Handbook of Experimental Pharmacology* 176:213-254.

- [46] Ogonowski AA, Kaesemeyer WH, Jin L, Ganapathy V, Leibach FH, Caldwell RW (2000) Effects of NO donors and synthase agonists on endothelial cell uptake of L-Arg and superoxide production. *American Journal of Physiology, Cell Physiology* 278:C136-C143.
- [47] Closs EI, Scheld JS, Sharafi M, Forstermann U (2000) Substrate supply for nitric-oxide synthase in macrophages and endothelial cells: role of cationic amino acid transporters. *Molecular Pharmacology* 57:68-74.
- [48] De Meyer GR, Herman AG (1997) Vascular endothelial dysfunction. *Progress in cardiovascular diseases* 39:325-342.
- [49] Deves R, Boyd CA (1998) Transporters for cationic amino acids in animal cells: discovery, structure, and function. *Physiological reviews* 78:487-545.
- [50] Verrey F, Closs EI, Wagner CA, Palacin M, Endou H, Kanai Y (2004) CATs and HATs: the SLC7 family of amino acid transporters. *European journal of physiology* 447:532-542.
- [51] Mann GE, Yudilevich DL, Sobrevia L (2003) Regulation of amino acid and glucose transporters in endothelial and smooth muscle cells. *Physiological reviews* 83:183-252.
- [52] Flores C, Rojas S, Aguayo C, Parodi J, Mann G, Pearson JD, et al. (2003) Rapid stimulation of L-arginine transport by D-glucose involves p42/44(mapk) and nitric oxide in human umbilical vein endothelium. *Circulation research* 92:64-72.
- [53] Vasquez G, Sanhueza F, Vasquez R, Gonzalez M, San Martin R, Casanello P, et al (2004) Role of adenosine transport in gestational diabetes-induced L-arginine transport and nitric oxide synthesis in human umbilical vein endothelium. *The journal of physiology* 560:111-122.
- [54] Gonzalez M, Gallardo V, Rodriguez N, Salomon C, Westermeier F, Guzman-Gutierrez E, et al. (2011) Insulin-stimulated L-arginine transport requires SLC7A1 gene expression and is associated with human umbilical vein relaxation. *Journal of cellular physiology* 226:2916-2924.
- [55] Cooke JP, Dzau VJ (1997) Nitric oxide synthase: role in the genesis of vascular disease. *Annual review of medicine* 48:489-509.
- [56] Shimokawa H (1999) Primary endothelial dysfunction: atherosclerosis. *Journal of molecular and cellular cardiology* 31:23-37.
- [57] Cai H, Harrison DG (2000) Endothelial dysfunction in cardiovascular diseases: the role of oxidant stress. *Circulation research* 87:840-844.
- [58] Davignon J, Ganz P (2004) Role of endothelial dysfunction in atherosclerosis. *Circulation* 109:27-32.

- [59] Walsh JH, Yong G, Cheetham C, Watts GF, O'Driscoll GJ, Taylor RR, et al. (2003) Effects of exercise training on conduit and resistance vessel function in treated and untreated hypercholesterolaemic subjects. *European heart journal* 24:1681-1689.
- [60] Ingram DG, Newcomer SC, Price EM, Eklund KE, McAllister RM, Laughlin MH (2007) Chronic nitric oxide synthase inhibition blunts endothelium-dependent function of conduit coronary arteries, not arterioles. *American journal of physiology - Heart and circulatory physiology* 292:H2798-H2808.
- [61] Tanner FC, Boulanger CM, Luscher TF (1993) Endothelium-derived nitric oxide, endothelin, and platelet vessel wall interaction: alterations in hypercholesterolemia and atherosclerosis. *Seminars In Thrombosis And Hemostasis* 19:167-175.
- [62] Mathew V, Cannan CR, Miller VM, Barber DA, Hasdai D, Schwartz RS, et al. (1997) Enhanced endothelin-mediated coronary vasoconstriction and attenuated basal nitric oxide activity in experimental hypercholesterolemia. *Circulation* 96:1930-1936.
- [63] Casino PR, Kilcoyne CM, Quyyumi AA, Hoeg JM, Panza JA (1993) The role of nitric oxide in endothelium-dependent vasodilation of hypercholesterolemic patients. *Circulation* 88:2541-2547.
- [64] Wennmalm A (1994) Nitric oxide (NO) in the cardiovascular system: role in atherosclerosis and hypercholesterolemia. *Blood Pressure* 3:279-282.
- [65] Laroia ST, Ganti AK, Laroia AT, Tendulkar KK (2003) Endothelium and the lipid metabolism: the current understanding. *International Journal of Cardiology* 88:1-9.
- [66] Napoli C, Ignarro LJ (2009) Nitric oxide and pathogenic mechanisms involved in the development of vascular diseases. *Archives of Pharmacal Research* 32:1103-1108.
- [67] Searle A, Gomez-Rosso L, Merono T, Salomon C, Duran-Sandoval D, Giunta G, et al. (2011) High LDL levels are associated with increased lipoprotein-associated phospholipase A(2) activity on nitric oxide synthesis and reactive oxygen species formation in human endothelial cells. *Clinical Biochemistry* 44:171-177.
- [68] Verbeuren TJ, Jordaens FH, Zonnekeyn LL, Van Hove CE, Coene MC, Herman AG (1986) Effect of hypercholesterolemia on vascular reactivity in the rabbit. I. Endothelium-dependent and endothelium-independent contractions and relaxations in isolated arteries of control and hypercholesterolemic rabbits. *Circulation research* 58:552-564.
- [69] Galle J, Busse R, Bassenge E (1991) Hypercholesterolemia and atherosclerosis change vascular reactivity in rabbits by different mechanisms. *Arteriosclerosis and thrombosis : a journal of vascular biology / American Heart Association* 11:1712-1718.
- [70] Gilligan DM, Guetta V, Panza JA, Garcia CE, Quyyumi AA, Cannon RO (1994) Selective loss of microvascular endothelial function in human hypercholesterolemia. *Circulation* 90:35-41.

- [71] Stapleton PA, Goodwill AG, James ME, Frisbee JC (2007) Altered mechanisms of endothelium-dependent dilation in skeletal muscle arterioles with genetic hypercholesterolemia. *American Journal of Physiology, Regulatory, Integrative and Comparative Physiology* 293:R1110-R1119.
- [72] Stapleton PA, Goodwill AG, James ME, Brock RW, Frisbee JC (2010) Hypercholesterolemia and microvascular dysfunction: interventional strategies. *Journal of Inflammation* 7:54.
- [73] Forstermann U, Mugge A, Alheid U, Haverich A, Frolich JC (1988) Selective attenuation of endothelium-mediated vasodilation in atherosclerotic human coronary arteries. *Circulation research* 62:185-190.
- [74] Posch K, Simecek S, Wascher TC, Jurgens G, Baumgartner-Parzer S, Kostner GM, et al. (1999) Glycated low-density lipoprotein attenuates shear stress-induced nitric oxide synthesis by inhibition of shear stress-activated L-arginine uptake in endothelial cells. *Diabetes* 48:1331-1337.
- [75] Jimenez A, Arriero MM, Lopez-Blaya A, Gonzalez-Fernandez F, Garcia R, Fortes J, et al. (2001) Regulation of endothelial nitric oxide synthase expression in the vascular wall and in mononuclear cells from hypercholesterolemic rabbits. *Circulation* 104:1822-1830.
- [76] Vidal F, Colome C, Martinez-Gonzalez J, Badimon L (1998) Atherogenic concentrations of native low-density lipoproteins down-regulate nitric-oxide-synthase mRNA and protein levels in endothelial cells. *European journal of biochemistry* 252:378-384.
- [77] Blair A, Shaul PW, Yuhanna IS, Conrad PA, Smart EJ (1999) Oxidized low density lipoprotein displaces endothelial nitric-oxide synthase (eNOS) from plasmalemmal caveolae and impairs eNOS activation. *The Journal of Biological Chemistry* 274:32512-32519.
- [78] Feron O, Dessy C, Moniotte S, Desager JP, Balligand JL (1999) Hypercholesterolemia decreases nitric oxide production by promoting the interaction of caveolin and endothelial nitric oxide synthase. *The Journal of clinical investigation* 103:897-905.
- [79] Everson WV, Smart EJ (2001) Influence of caveolin, cholesterol, and lipoproteins on nitric oxide synthase: implications for vascular disease. *Trends in Cardiovascular Medicine* 11:246-250.
- [80] Feron O, Dessy C, Desager JP, Balligand JL (2001) Hydroxy-methylglutaryl-coenzyme A reductase inhibition promotes endothelial nitric oxide synthase activation through a decrease in caveolin abundance. *Circulation* 103:113-118.
- [81] Shaul PW (2002) Regulation of endothelial nitric oxide synthase: location, location, location. *Annual Review of Physiology* 64:749-774.

- [82] Zhu Y, Liao HL, Niu XL, Yuan Y, Lin T, Verna L, et al. (2003) Low density lipoprotein induces eNOS translocation to membrane caveolae: the role of RhoA activation and stress fiber formation. *Biochimica et Biophysica Acta* 1635:117-126.
- [83] Michel T, Vanhoutte PM (2010) Cellular signaling and NO production. *Pflügers archiv - European journal of physiology* 459:807-816.
- [84] Garcia-Cardena G, Fan R, Stern DF, Liu J, Sessa WC (1996) Endothelial nitric oxide synthase is regulated by tyrosine phosphorylation and interacts with caveolin-1. *The Journal of Biological Chemistry* 271:27237-27240.
- [85] Shaul PW, Smart EJ, Robinson LJ, German Z, Yuhanna IS, Ying Y, et al. (1996) Acylation targets endothelial nitric-oxide synthase to plasmalemmal caveolae. *The Journal of Biological Chemistry* 271:6518-6522.
- [86] Liu J, Garcia-Cardena G, Sessa WC (1996) Palmitoylation of endothelial nitric oxide synthase is necessary for optimal stimulated release of nitric oxide: implications for caveolae localization. *Biochemistry* 35:13277-13281.
- [87] Michel JB, Feron O, Sacks D, Michel T (1997) Reciprocal regulation of endothelial nitric-oxide synthase by Ca<sup>2+</sup>-calmodulin and caveolin. *The Journal of Biological Chemistry* 272:15583-15586.
- [88] Bist A, Fielding PE, Fielding CJ (1997) Two sterol regulatory element-like sequences mediate up-regulation of caveolin gene transcription in response to low density lipoprotein free cholesterol. *Proceedings of the National Academy of Sciences U S A* 94:10693-10698.
- [89] Fielding CJ, Bist A, Fielding PE (1997) Caveolin mRNA levels are up-regulated by free cholesterol and down-regulated by oxysterols in fibroblast monolayers. *Proceedings of the National Academy of Sciences U S A* 94:3753-3758.
- [90] Hailstones D, Slier LS, Parton RG, Stanley KK (1998) Regulation of caveolin and caveolae by cholesterol in MDCK cells. *The Journal of Lipid Research* 39:369-379.
- [91] Boger RH, Bode-Boger SM, Szuba A, Tsao PS, Chan JR, Tangphao O, et al. (1998) Asymmetric dimethylarginine (ADMA): a novel risk factor for endothelial dysfunction: its role in hypercholesterolemia. *Circulation* 98:1842-1847.
- [92] Boger RH, Bode-Boger SM (2000) Asymmetric dimethylarginine, derangements of the endothelial nitric oxide synthase pathway, and cardiovascular diseases. *Seminars In Thrombosis And Hemostasis* 26:539-545.
- [93] White V, Gonzalez E, Capobianco E, Pustovrh C, Sonez C, Romanini MC, et al. (2004) Modulatory effect of leptin on nitric oxide production and lipid metabolism in term placental tissues from control and streptozotocin-induced diabetic rats. *Reproduction, Fertility and Development* 16:363-372.
- [94] Vladimirova-Kitova L, Deneva T, Angelova E, Nikolov F, Marinov B, Mateva N (2008) Relationship of asymmetric dimethylarginine with flow-mediated dilatation in



subjects with newly detected severe hypercholesterolemia. *Clinical Physiology and Functional Imaging* 28:417-425.

- [95] Leiper J, Nandi M (2011) The therapeutic potential of targeting endogenous inhibitors of nitric oxide synthesis. *Nature Reviews Drug Discovery* 10:277-291.
- [96] Boger RH, Sydow K, Borlak J, Thum T, Lenzen H, Schubert B, et al. (2000) LDL cholesterol upregulates synthesis of asymmetrical dimethylarginine in human endothelial cells: involvement of S-adenosylmethionine-dependent methyltransferases. *Circulation research* 87:99-105.
- [97] Ito A, Tsao PS, Adimoolam S, Kimoto M, Ogawa T, Cooke JP (1999) Novel mechanism for endothelial dysfunction: dysregulation of dimethylarginine dimethylaminohydrolase. *Circulation* 99:3092-3095.
- [98] Kawashima S, Yokoyama M (2004) Dysfunction of endothelial nitric oxide synthase and atherosclerosis. *Arteriosclerosis, Thrombosis, and Vascular Biology* 24:998-1005.
- [99] Ozaki M, Kawashima S, Yamashita T, Hirase T, Namiki M, Inoue N, et al. (2002) Overexpression of endothelial nitric oxide synthase accelerates atherosclerotic lesion formation in apoE-deficient mice. *The Journal of clinical investigation* 110:331-340.
- [100] Tang FT, Qian ZY, Liu PQ, Zheng SG, He SY, Bao LP, et al. (2006) Crocetin improves endothelium-dependent relaxation of thoracic aorta in hypercholesterolemic rabbit by increasing eNOS activity. *Biochemical Pharmacology* 72:558-565.
- [101] Stroes E, Kastelein J, Cosentino F, Erkelens W, Wever R, Koomans H, et al. (1997) Tetrahydrobiopterin restores endothelial function in hypercholesterolemia. *The Journal of clinical investigation* 99:41-46.
- [102] Tiefenbacher CP, Bleeke T, Vahl C, Amann K, Vogt A, Kubler W (2000) Endothelial dysfunction of coronary resistance arteries is improved by tetrahydrobiopterin in atherosclerosis. *Circulation* 102:2172-2179.
- [103] Tachibana D, Fukumasu H, Shintaku H, Fukumasu Y, Yamamasu S, Ishiko O, et al. (2002) Decreased plasma tetrahydrobiopterin in pregnant women is caused by impaired 6-pyruvoyl tetrahydropterin synthase activity. *International Journal of Molecular Medicine* 9:49-52.
- [104] Iwanaga N, Yamamasu S, Tachibana D, Nishio J, Nakai Y, Shintaku H, et al. (2004) Activity of synthetic enzymes of tetrahydrobiopterin in the human placenta. *International Journal of Molecular Medicine* 13:117-120.
- [105] Dulak J, Polus M, Guevara I, Hartwich J, Wybranska I, Krzesz R, et al. (1999) Oxidized low density lipoprotein inhibits inducible nitric oxide synthase, GTP cyclohydrolase I and transforming growth factor beta gene expression in rat macrophages. *Journal of Physiology and Pharmacology* 50:429-441.
- [106] Dulak J, Polus M, Guevara I, Polus A, Hartwich J, Dembinska-Kiec A (1997) Regulation of inducible nitric oxide synthase (iNOS) and GTP cyclohydrolase I (GTP-CH I)

gene expression by ox-LDL in rat vascular smooth muscle cells. *Journal of Physiology and Pharmacology* 48:689-697.

- [107] Gonzalez M, Flores C, Pearson JD, Casanello P, Sobrevia L (2004) Cell signalling-mediated insulin increase of mRNA expression for cationic amino acid transporters-1 and -2 and membrane hyperpolarization in human umbilical vein endothelial cells. *Pflügers archiv - European journal of physiology* 448:383-394.
- [108] González M, Puebla C, Guzmán-Gutierrez E, Cifuentes F, Nien J, Abarzua F, et al. (2011) Maternal and fetal metabolic dysfunction in pregnancy diseases associated with vascular oxidative and nitrosative stress. Book: the molecular basis for the link between maternal health and the origin of the fetal congenital abnormalities 8:98-115.
- [109] Jay MT, Chirico S, Siow RC, Bruckdorfer KR, Jacobs M, Leake DS, et al. (1997) Modulation of vascular tone by low density lipoproteins: effects on L-arginine transport and nitric oxide synthesis. *Experimental Physiology* 82:349-360.
- [110] Nuzkowski A, Grabner R, Marsche G, Unbehaun A, Malle E, Heller R (2001) Hypochlorite-modified low density lipoprotein inhibits nitric oxide synthesis in endothelial cells via an intracellular dislocalization of endothelial nitric-oxide synthase. *The Journal of Biological Chemistry* 276:14212-14221.
- [111] Kikuta K, Sawamura T, Miwa S, Hashimoto N, Masaki T (1998) High-affinity arginine transport of bovine aortic endothelial cells is impaired by lysophosphatidylcholine. *Circulation research* 83:1088-1096.
- [112] Vergnani L, Hatric S, Ricci F, Passaro A, Manzoli N, Zuliani G, et al. (2000) Effect of native and oxidized low-density lipoprotein on endothelial nitric oxide and superoxide production : key role of L-arginine availability. *Circulation* 101:1261-1266.
- [113] Zhang WZ, Venardos K, Finch S, Kaye DM (2008) Detrimental effect of oxidized LDL on endothelial arginine metabolism and transportation. *The International Journal of Biochemistry and Cell Biology* 40:920-928.
- [114] Ryoo S, Bhunia A, Chang F, Shoukas A, Berkowitz DE, Romer LH (2011) OxLDL-dependent activation of arginase II is dependent on the LOX-1 receptor and downstream RhoA signaling. *Atherosclerosis* 214:279-287.
- [115] Ryoo S, Gupta G, Benjo A, Lim HK, Camara A, Sikka G, et al. (2008) Endothelial arginase II: a novel target for the treatment of atherosclerosis. *Circulation research* 102:923-932.
- [116] Erdely A, Kepka-Lenhart D, Salmen-Muniz R, Chapman R, Hulderman T, Kashon M, et al. (2010) Arginase activities and global arginine bioavailability in wild-type and ApoE-deficient mice: responses to high fat and high cholesterol diets. *PLoS One* 5:e15253.

- [117] Holowatz LA, Santhanam L, Webb A, Berkowitz DE, Kenney WL (2011) Oral atorvastatin therapy restores cutaneous microvascular function by decreasing arginase activity in hypercholesterolaemic humans. *The journal of physiology* 589:2093-2103.
- [118] Metzger BE, Coustan DR (1998) Summary and recommendations of the Fourth International Workshop-Conference on Gestational Diabetes Mellitus. The Organizing Committee. *Diabetes Care* 21:S161-S167.
- [119] Metzger BE, Buchanan TA, Coustan DR, de Leiva A, Dunger DB, Hadden DR, et al. (2007) Summary and recommendations of the Fifth International Workshop-Conference on Gestational Diabetes Mellitus. *Diabetes Care* 30:S251-S260.
- [120] American Diabetes Association (2011) Diagnosis and classification of diabetes mellitus. *Diabetes Care* 34:S62-S69.
- [121] Nold JL, Georgieff MK (2004) Infants of diabetic mothers. *Pediatric Clinics of North America* 51:619-637.
- [122] Greene MF, Solomon CG (2005) Gestational diabetes mellitus -- time to treat. *The New England Journal of Medicine* 352:2544-2546.
- [123] Desoye G, Hauguel-de Mouzon S (2007) The human placenta in gestational diabetes mellitus. The insulin and cytokine network. *Diabetes Care* 33:S120-S126.
- [124] Belmar J C, Salinas C P, Becker V J, Abarzúa C F, Olmos C P, González B P, et al. (2004) Incidencia de diabetes gestacional segun distintos metodos diagnosticos y sus implicancias clinicas. *Revista Chilena de Obstetricia y Ginecología* 69:2-7.
- [125] Huidobro A, Fulford A, Carrasco E (2004) Incidence of gestational diabetes and relationship to obesity in Chilean pregnant women. *Revista Medica de Chile* 132:931-938.
- [126] Ferrara A, Kahn HS, Quesenberry CP, Riley C, Hedderson MM (2004) An increase in the incidence of gestational diabetes mellitus: Northern California, 1991-2000. *Obstetrics and Gynecology* 103:526-533.
- [127] Dabelea D, Snell-Bergeon JK, Hartsfield CL, Bischoff KJ, Hamman RF, McDuffie RS (2005) Increasing prevalence of gestational diabetes mellitus (GDM) over time and by birth cohort: Kaiser Permanente of Colorado GDM Screening Program. *Diabetes Care* 28:579-584.
- [128] Robitaille J, Grant AM (2008) The genetics of gestational diabetes mellitus: evidence for relationship with type 2 diabetes mellitus. *Genetics in Medicine* 10:240-250.
- [129] Omar HA, Ramirez R, Arsich J, Tracy T, Glover D, Gibson M (1998) Reduction of the Human Placental Vascular Relaxation to Progesterone by Gestational Diabetes. *Journal of Maternal-Fetal Investigation* 8:27-30.
- [130] De Vriese AS, Verbeuren TJ, Van de Voorde J, Lameire NH, Vanhoutte PM (2000) Endothelial dysfunction in diabetes. *British Journal of Pharmacology* 130:963-974.

- [131] Tchirikov M, Rybakowski C, Huneke B, Schoder V, Schroder HJ (2002) Umbilical vein blood volume flow rate and umbilical artery pulsatility as 'venous-arterial index' in the prediction of neonatal compromise. *Ultrasound Obstetrics and Gynecology* 20:580-585.
- [132] Biri A, Onan A, Devrim E, Babacan F, Kavutcu M, Durak I (2006) Oxidant status in maternal and cord plasma and placental tissue in gestational diabetes. *Placenta* 27:327-332.
- [133] Guzman-Gutierrez E, Westermeier F, Salomon C, Gonzalez M, Pardo F, Leiva A, et al. (2012) Insulin-Increased L-Arginine Transport Requires A(2A) Adenosine Receptors Activation in Human Umbilical Vein Endothelium. *PLoS One* 7:e41705.
- [134] Salomon C, Westermeier F, Puebla C, Arroyo P, Guzman-Gutierrez E, Pardo F, et al. (2012) Gestational diabetes reduces adenosine transport in human placental microvascular endothelium, an effect reversed by insulin. *PLoS One* 7:e40578.
- [135] von Mandach U, Lauth D, Huch R (2003) Maternal and fetal nitric oxide production in normal and abnormal pregnancy. *Journal of Maternal-Fetal and Neonatal Medicine* 13:22-27.
- [136] Farias M, San Martin R, Puebla C, Pearson JD, Casado JF, Pastor-Anglada M, et al. (2006) Nitric oxide reduces adenosine transporter ENT1 gene (SLC29A1) promoter activity in human fetal endothelium from gestational diabetes. *Journal of cellular physiology* 208:451-460.
- [137] National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) (2002) Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. *Circulation* 106:3143-3421.
- [138] Roger VL, Go AS, Lloyd-Jones DM, Adams RJ, Berry JD, Brown TM, et al. (2011) Heart Disease and Stroke Statistics--2011 Update: A Report From the American Heart Association. *Circulation* 123:e18-e209.
- [139] Arsenault BJ, Boekholdt SM, Kastelein JJ (2011) Lipid parameters for measuring risk of cardiovascular disease. *Nature Reviews Cardiology* 8:197-206.
- [140] Boney CM, Verma A, Tucker R, Vohr BR (2005) Metabolic syndrome in childhood: association with birth weight, maternal obesity, and gestational diabetes mellitus. *Pediatrics* 115:e290-e296.
- [141] Clausen TD, Mathiesen ER, Hansen T, Pedersen O, Jensen DM, Lauenborg J, et al. (2009) Overweight and the metabolic syndrome in adult offspring of women with diet-treated gestational diabetes mellitus or type 1 diabetes. *The Journal of Clinical Endocrinology and Metabolism* 94:2464-2470.

- [142] Moore TR (2010) Fetal exposure to gestational diabetes contributes to subsequent adult metabolic syndrome. *American Journal of Obstetrics and Gynecology* 202:643-649.
- [143] Isomaa B, Almgren P, Tuomi T, Forsen B, Lahti K, Nissen M, et al. (2001) Cardiovascular morbidity and mortality associated with the metabolic syndrome. *Diabetes Care* 24:683-689.
- [144] Egeland GM, Meltzer SJ. (2010) Following in mother's footsteps? Mother-daughter risks for insulin resistance and cardiovascular disease 15 years after gestational diabetes. *Diabetic Medicine* 27:257-265.
- [145] Pirkola J, Vaarasmaki M, Ala-Korpela M, Bloigu A, Canoy D, Hartikainen AL, et al. (2010) Low-grade, systemic inflammation in adolescents: association with early-life factors, gender, and lifestyle. *American Journal of Epidemiology* 171:72-82.
- [146] Mottillo S, Filion KB, Genest J, Joseph L, Pilote L, Poirier P, et al. (2010) The metabolic syndrome and cardiovascular risk a systematic review and meta-analysis. *Journal of the American College of Cardiology* 56:1113-1132.
- [147] Sanchez-Vera I, Bonet B, Viana M, Quintanar A, Martin MD, Blanco P, et al. (2007) Changes in plasma lipids and increased low-density lipoprotein susceptibility to oxidation in pregnancies complicated by gestational diabetes: consequences of obesity. *Metabolism* 56:1527-1533.
- [148] Zawiejska A, Wender-Ozegowska E, Brazert J, Sadowski K (2008) Components of metabolic syndrome and their impact on fetal growth in women with gestational diabetes mellitus. *Journal of Physiology and Pharmacology* 59:5-18.
- [149] Reece E. (2010) The fetal and maternal consequences of gestational diabetes mellitus. *Journal of Maternal-Fetal and Neonatal Medicine* 23:199-203.
- [150] Radaelli T, Lepercq J, Varastehpour A, Basu S, Catalano PM, Hauguel-De Mouzon S (2009) Differential regulation of genes for fetoplacental lipid pathways in pregnancy with gestational and type 1 diabetes mellitus. *American Journal of Obstetrics and Gynecology* 201:209 e201-209 e210.
- [151] Scholler M, Wadsack C, Lang I, Etschmaier K, Schweinzer C, Marsche G, et al. (2012) Phospholipid transfer protein in the placental endothelium is affected by gestational diabetes mellitus. *The Journal of Clinical Endocrinology and Metabolism* 97:437-445.
- [152] Marceau G, Volle DH, Gallot D, Mangelsdorf DJ, Sapin V, Lobaccaro JM (2005) Placental expression of the nuclear receptors for oxysterols LXRalpha and LXRbeta during mouse and human development. *The anatomical record. Part A, Discoveries in molecular, cellular, and evolutionary biology* 283:175-181.
- [153] Scholler M, Wadsack C, Metso J, Chirackal Manavalan AP, Sreckovic I, Schweinzer C, et al. (2012) Phospholipid Transfer Protein Is Differentially Expressed in Human

Arterial and Venous Placental Endothelial Cells and Enhances Cholesterol Efflux to Fetal HDL. *The Journal of Clinical Endocrinology and Metabolism* 97:2466-2474.

- [154] Ferderbar S, Pereira EC, Apolinario E, Bertolami MC, Faludi A, Monte O, et al. (2007) Cholesterol oxides as biomarkers of oxidative stress in type 1 and type 2 diabetes mellitus. *Diabetes/Metabolism Research and Reviews* 23:35-42.
- [155] Skilton MR (2008) Intrauterine risk factors for precocious atherosclerosis. *Pediatrics* 121:570-574.
- [156] Sarikabadayi YU, Aydemir O, Kanmaz G, Aydemir C, Oguz SS, Erdeve O, et al. (2012) Umbilical artery intima-media and wall thickness in infants of diabetic mothers. *Neonatology* 102:157-162.
- [157] Vahratian A, Misra VK, Trudeau S, Misra DP (2010) Prepregnancy body mass index and gestational age-dependent changes in lipid levels during pregnancy. *Obstetrics and Gynecology* 116:107-113.
- [158] Knopp RH, Van Allen MI, McNeely M, Walden CE, Plovie B, Shiota K, et al. (1993) Effect of insulin-dependent diabetes on plasma lipoproteins in diabetic pregnancy. *The Journal of Reproductive Medicine* 38:703-710.
- [159] Vanderjagt DJ, Patel RJ, El-Nafaty AU, Melah GS, Crossey MJ, Glew RH (2004) High-density lipoprotein and homocysteine levels correlate inversely in preeclamptic women in northern Nigeria. *Acta Obstetrica et Gynecologica Scandinavica* 83:536-542.
- [160] Ywaskewycz L BG, Castillo MS, López D, Pedrozo W (2010) Perfil lipídico por trimestre de gestación en una población de mujeres adultas. *Revista Chilena de Obstetricia y Ginecología* 75:227-233.
- [161] Sattar N, Greer IA, Loudon J, Lindsay G, McConnell M, Shepherd J, et al. (1997) Lipoprotein subfraction changes in normal pregnancy: threshold effect of plasma triglyceride on appearance of small, dense low density lipoprotein. *The Journal of Clinical Endocrinology and Metabolism* 82:2483-2491.
- [162] Martin U, Davies C, Hayavi S, Hartland A, Dunne F (1999) Is normal pregnancy atherogenic? *Clinical Science* 96:421-425.
- [163] Koukkou E, Watts GF, Mazurkiewicz J, Lowy C (1994) Ethnic differences in lipid and lipoprotein metabolism in pregnant women of African and Caucasian origin. *Journal of Clinical Pathology* 47:1105-1107.
- [164] Mazurkiewicz JC, Watts GF, Warburton FG, Slavin BM, Lowy C, Koukkou E (1994) Serum lipids, lipoproteins and apolipoproteins in pregnant non-diabetic patients. *Journal of Clinical Pathology* 47:728-731.
- [165] Liguori A, D'Armiento FP, Palagiano A, Balestrieri ML, Williams-Ignarro S, de Nigris F, et al. (2007) Effect of gestational hypercholesterolaemia on omental vasoreactivity,

- placental enzyme activity and transplacental passage of normal and oxidised fatty acids. *BJOG: An International Journal of Obstetrics and Gynaecology* 114:1547-1556.
- [166] Magnussen EB, Vatten LJ, Smith GD, Romundstad PR (2009) Hypertensive disorders in pregnancy and subsequently measured cardiovascular risk factors. *Obstetrics and Gynecology* 114:961-970.
- [167] Bon C, Raudrant D, Golfier F, Poloce F, Champion F, Pichot J, et al. (2007) [Feto-maternal metabolism in human normal pregnancies: study of 73 cases]. *Annales de biologie clinique* 65:609-619.
- [168] Schaefer-Graf UM, Graf K, Kulbacka I, Kjos SL, Dudenhausen J, Vetter K, et al. (2008) Maternal lipids as strong determinants of fetal environment and growth in pregnancies with gestational diabetes mellitus. *Diabetes Care* 31:1858-1863.
- [169] Ordovas JM, Pocovi M, Grande F (1984) Plasma lipids and cholesterol esterification rate during pregnancy. *Obstetrics and Gynecology* 63:20-25.
- [170] Belo L, Caslake M, Gaffney D, Santos-Silva A, Pereira-Leite L, Quintanilha A, et al. (2002) Changes in LDL size and HDL concentration in normal and preeclamptic pregnancies. *Atherosclerosis* 162:425-432.
- [171] Saarelainen H, Laitinen T, Raitakari OT, Juonala M, Heiskanen N, Lyyra-Laitinen T, et al. (2006) Pregnancy-related hyperlipidemia and endothelial function in healthy women. *Circulation Journal* 70:768-772.
- [172] Ogura K, Miyatake T, Fukui O, Nakamura T, Kameda T, Yoshino G (2002) Low-density lipoprotein particle diameter in normal pregnancy and preeclampsia. *Journal of Atherosclerosis and Thrombosis* 9:42-47.
- [173] Ouyang Y, Chen H, Chen H (2007) Reduced plasma adiponectin and elevated leptin in pre-eclampsia. *International Journal of Gynecology and Obstetrics* 98:110-114.
- [174] Iftikhar U, Iqbal A, Shakoor S (2010) Relationship between leptin and lipids during pre-eclampsia. *Journal of the Pakistan Medical Association* 60:432-435.
- [175] Mshelia DS, Kullima A, Gali RM, Kawuwa MB, Mamza YP, Habu SA, et al. (2010) The use of plasma lipid and lipoprotein ratios in interpreting the hyperlipidaemia of pregnancy. *Journal of obstetrics and gynaecology* 30:804-808.
- [176] Grissa O, Ategbro JM, Yessoufou A, Tabka Z, Miled A, Jerbi M, et al. (2007) Antioxidant status and circulating lipids are altered in human gestational diabetes and macrosomia. *Translational Research* 150:164-171.
- [177] Rossmannith WG, Hoffmeister U, Wolfahrt S, Kleine B, McLean M, Jacobs RA, et al. (1999) Expression and functional analysis of endothelial nitric oxide synthase (eNOS) in human placenta. *Molecular Human Reproduction* 5:487-494.
- [178] Di Iulio JL, Gude NM, King RG, Li CG, Rand MJ, Brennecke SP (1999) Human placental nitric oxide synthase activity is not altered in diabetes. *Clinical Science* 97:123-128.

- [179] Thornburg KL, O'Tierney PF, Louey S (2010) Review: The placenta is a programming agent for cardiovascular disease. *Placenta* 31:S54-S59.
- [180] Lee MY, Cai Y, Wang Y, Liao SY, Liu Y, Zhang Y, et al. (2012) Differential genomic changes caused by cholesterol- and PUFA-rich diets in regenerated porcine coronary endothelial cells. *Physiological Genomics* 44:551-561.
- [181] Chan E, Chan JY, Wu JH, Wan CW, Leung GP, Lee SM, et al. (2012) Serum nitric oxide synthase activity is a novel predictor of impaired vasorelaxation in rats. *Clinical and Experimental Pharmacology and Physiology* 39:894-896.
- [182] Kim OY, Lee SM, Chung JH, Do HJ, Moon J, Shin MJ (2012) Arginase I and the very low-density lipoprotein receptor are associated with phenotypic biomarkers for obesity. *Nutrition* 28:635-639.
- [183] Hollingsworth DR, Grundy SM (1982) Pregnancy-associated hypertriglyceridemia in normal and diabetic women. Differences in insulin-dependent, non-insulin-dependent, and gestational diabetes. *Diabetes* 31:1092-1097.
- [184] Tarim E, Yigit F, Kilicdag E, Bagis T, Demircan S, Simsek E, et al. (2006) Early onset of subclinical atherosclerosis in women with gestational diabetes mellitus. *Ultrasound Obstetrics and Gynecology* 27:177-182.
- [185] Koukkou E, Watts GF, Lowy C (1996) Serum lipid, lipoprotein and apolipoprotein changes in gestational diabetes mellitus: a cross-sectional and prospective study. *Journal of Clinical Pathology* 49:634-637.
- [186] Knopp RH, LaRosa JC, Burkman RT (1993) Contraception and dyslipidemia. *American Journal of Obstetrics and Gynecology* 168:1994-2005.
- [187] Rizzo M, Berneis K, Altinova AE, Toruner FB, Akturk M, Ayvaz G, et al. (2008) Atherogenic lipoprotein phenotype and LDL size and subclasses in women with gestational diabetes. *Diabetic Medicine* 25:1406-1411.

IntechOpen



