Diplomarbeit

The Effects of Smoking on Preeclampsia: A Systematic Review

eingereicht von

Sebastian Lind

zur Erlangung des akademischen Grades

Doktor der gesamten Heilkunde

(Dr. med. univ.)

an der

Medizinischen Universität Graz

ausgeführt am

Lehrstuhl für Physiologie

unter der Anleitung von

Assoz. Prof. Priv.-Doz. Dr.med. MMedSci PhD Nandu Goswami

Assoz. Univ.-Profin. Dr.med. Mila Cervar-Zivkovic

Graz, am 11.05.2022

Eidesstattliche Erklärung

Ich erkläre ehrenwörtlich, dass ich die vorliegende Arbeit selbstständig und ohne fremde Hilfe verfasst habe, andere als die angegebenen Quellen nicht verwendet habe und die den benutzten Quellen wörtlich oder inhaltlich entnommenen Stellen als solche kenntlich gemacht habe.

Graz, am 11.05.2022 Sebastian Lind eh

Table of Contents

1. Glossary and Abbreviations

ACE	angiotensinogen converting enzyme
ADH	antidiuretic hormone
Ang-1	angiopoietin 1
ANP	atrial natriuretic peptide
AT1	angiotensin I
AT1-AAs	angiotensin II type 1 receptor agonistic antibodies
AT2	angiotensin II
BWGA	birth weight for gestational age
cGMP	cyclic guanosine monophosphate
COX	cyclooxygenase
dpf	day post fertilization
EDHF	endothelium-derived hyperpolarizing factor
eEVT	endovascular extravillous cells
GFR	glomerular filtration rate
GH	gestational hypertension
hCG	human chorionic gonadotropin
HDP	hypertensive disorders of pregnancy
iEVT	interstitial extravillous cells
IL-1B	interleukin-1B
LBW	low birth weight
nAChR	nicotinic acetylcholine receptors
NaCl	sodium chloride
NK	natural killer
NO	nitric oxide
NOS3	nitric oxide synthase 3

PAPP-A	pregnancy-associated plasma protein A
PE	preeclampsia
PGE	prostaglandin E
PGE2	prostaglandin E2
PGI2	prostacyclin
PlGF	placental growth factor
PPE	preterm preeclampsia
RAAS	renin angiotensin aldosterone system
sEng	soluble endoglin
sFLT-1	soluble fms-like tyrosine kinase-1
SGA	small for gestational age
SMC	smooth muscle cell
SMCs	smooth muscle cells
TE	trophectoderm
V1	vasopressin receptor 1
V2	vasopressin receptor 2
VEGF	vascular endothelial growth factor
VSMC	vascular smooth muscle cell
NH	non-hispanic

2. List of figures and tables

List of Figures

List of tables

3. Zusammenfassung

Hintergrund: Präeklampsie ist eine der häufigsten Todesursachen für den Tod einer werdenden Mutter, und die Geburt ist noch immer ist die einzige kausale Therapie dieser Erkrankung. Während Rauchen für seine vielen gesundheitsschädlichen Effekte bekannt ist, eine der häufigsten Todesursachen der Welt ist, und insbesondere in der Schwangerschaft dringend davon abgeraten werden sollte, hat man einen schützenden Effekt bezüglich der Entwicklung von Präeklampsie beobachtet.

Ziele: Das Ziel diese Diplomarbeit ist die gegenwärtige Literatur zu diesem Thema systematisch zu untersuchen. Die gefundene Literatur wurde in vier Kategorien unterteilt. Die erste soll einen Überblick über die Effekte des Rauchens bezüglich des reduzierten Risikos an Präeklampsie zu erleiden geben. Die zweite Kategorie untersucht die zugrundeliegenden Prozesse des Rauchens in Präeklampsie und zeigt die wichtigsten auf, um die Pathophysiologie der Erkrankung besser zu verstehen und potenzielle Therapieoptionen aufzuzeigen. Die letzten zwei Kategorien untersuchen mögliche Verzerrungen der Studien.

Methoden: Um die aktuelle Literatur zu diesem Thema zu finden wurde eine systemische Suche von Literatur Datenbanken durchgeführt. Hierbei wurden PubMed und Web of Science durchsucht, sowie die gefundene Literatur auf Sekundärliteratur durchsucht. Weiters wurden Informationen durch Bücher und E-Books sowie Publikationen durchsucht, die von der Bibliothek der Medizinischen Universität Graz zur Verfügung gestellt worden sind, Publikationen von PubMed, Google Scholar und Web of Science, Literatur und Leitlinien der World Health Organisation, der International Society for the Study of Hypertension in Pregnancy, sowie der Austrian, Swiss and German society of Gynecology and Obstetrics.

Ergebnisse und Diskussion: Schließlich wurden 27 Publikationen gefunden. Die untersuchte Literatur zeigte einen Trend zur Reduktion des Risikos von Präeklampsie durch Rauchen. Die Mechanismen dahinter waren unter anderem Veränderungen von PlGF, VGIF, sFlt-1, AM, B-HC und nAChR Rezeptor Untereinheiten Expression. Weiters wurde gezeigt, dass CO Effekte von PE verhindern konnte, Hypertension und Proteinurie verhindern konnte, den Umbau von Spiralarterien sowie die Durchblutung des Uterus gefördert hat in PE/HT Mäusen, Nikotin verminderte sFlt-1 und erhöhte VEGF in hypoxischen Zellkulturen. Studienverzerrungen scheinen plausibel, wie stark der Effekt ausgeprägt ist sollte jedoch weiter untersucht werden.

4. Abstract

Background: Preeclampsia is a leading cause of maternal death, and still, the only causal therapy for it is giving birth. And while smoking is known to have a lot of harmful effects on the human body, to be a main cause of death worldwide, and to be especially harmful in pregnant mothers, studies show that it has a protective effect on preeclampsia.

Aims and objectives: The aim of this thesis is to review the current literature given on this topic. The literature found is categorized in four different parts. The first one aims to give an overview of the effects of smoking regarding the risk reduction of preeclampsia. The second part reviews the underlying processes of smoking in preeclampsia to find out which ones are the most relevant and could be important for understanding the pathophysiology of the disease better and for finding possible therapy options. The last two parts investigate possible bias seen in the studies conducted on this topic.

Methodology: To review the current literature on this topic a systemic search of literature databases was executed. Databases searched were PubMed and Web of Science, and the found literature was then screened for secondary literature. Further information was gathered via books, e-Books and papers provided by the library of the Medical University of Graz, publications found via PubMed, Google Scholar and Web of Science, literature and guidelines provided by the World Health Organization, International Society for the Study of Hypertension in Pregnancy, the Austrian, Swiss and German society of Gynecology and Obstetrics.

Results and discussion: Eventually 27 publications were identified. The reviewed literature showed a general trend towards reducing the risk of preeclampsia due to smoking. The underlying mechanisms include alterations of PlGF, VGIF, sFlt-1, AM, B-HC and nAChR receptor subunit expression. Also, CO could prevent effects of PE, prevent HT, proteinuria, increase SA remodeling and increase uterine blood flow in PE/HT mice, and nicotine decreased s-Flt-1 and increased VEGF in hypoxic cell cultures. Study bias seems possible, but the impact must be further investigated.

5. Introduction

Being pregnant is a highly complicated process for a female body. To ensure that the foetus is supplied sufficiently with all of its necessities, an adequate uteroplacental blood flow is crucial. This uteroplacental blood flow is unfortunately disturbed by women suffering from preeclampsia, a hypertensive disease in pregnant woman. While there is a prospective study going on about the hemodynamic differences in preeclamptic women, conducted by the department of Physiology in Graz, this literature research aims to discuss all of the information available regarding this topic. In the introduction, an overview on the physiological and anatomical adaptations of a regular pregnancy will be given to better understand the mechanics of this disease. Then preeclampsia, the pathogenesis, the symptoms, and the possible therapies to this day will be described, and afterwards the effects of smoking in general and in pregnancy.

5.1. Pregnancy

5.1.1. Adaptation

A woman's body undergoes several drastic physiological changes to cope adequately with a pregnancy. There are changes described in the cardiovascular system, the immune system, the hematologic system as well as the intestines and many more. The most important changes regarding preeclampsia will be explained in this chapter.

5.1.1.1. Cardiovascular System

The cardiovascular system changes in a number of different ways. The cardiac output increases by 20% in eight weeks of gestation. This effect is mostly due to blood pressure regulation and endothelium dependent factors which will be explained in more detail later. The blood volume increases by 20%-100% in normal pregnancies, with an average of 45%. These changes are crucial for the systemic and uteroplacental blood flow, and therefore crucial for the normal development and nutrition of the child. (1) The heart rate increases continuously and reaches 80 beats/min in the third trimester, which is a plus of 12 b/m over the pregnancy.

5.1.1.2. Blood Pressure Regulation

The blood pressure usually goes down by around 4 mmHg during pregnancy but this differs depending on the stage of the pregnancy.(2) In preeclampsia the systolic blood pressure rises above 140 mmHg.

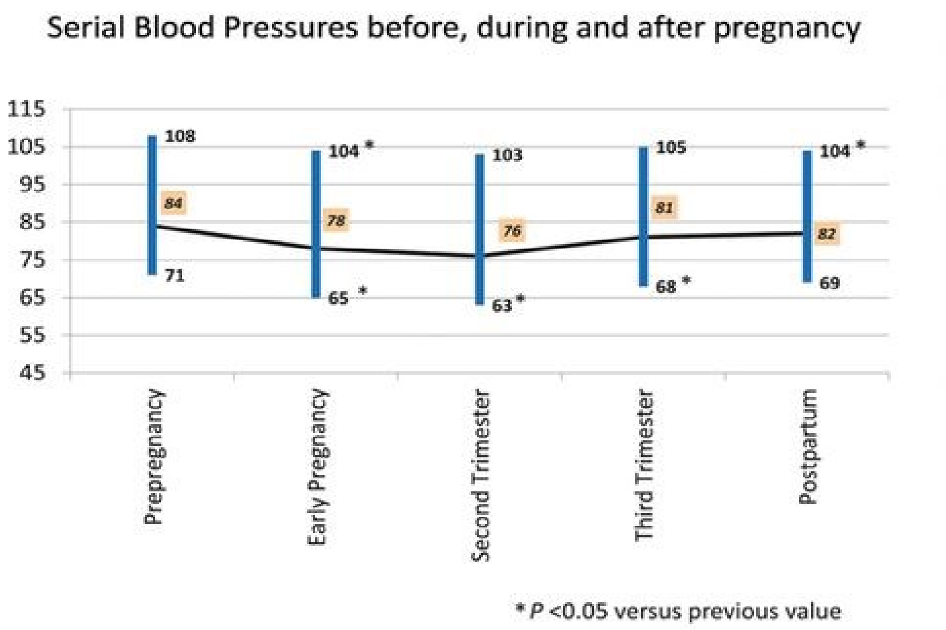


Figure 1: Serial Blood Pressure before, during and after pregnancy

(Reproduced from Mahendru AA. A Longitudinal Study of Maternal Cardiovascular Function from Preconception to the Postpartum Period. Journal of Hypertension. 2014 Apr;32(4):849-56,.)

Blood pressure regulation is an important factor for the physiologic process of the pregnancy. The main factors in the regulation of blood pressure are viscosity, vessel diameter, and elasticity of the vessels. (3) For a better understanding, these factors will be explained in more detail, as well as the adaptations made in a physiological pregnancy.

Short-term blood pressure regulation is done by the baroreceptors in the aorta and in the carotid artery, as well as the volume receptors in the right atrium. In the case of sensing a high blood pressure through stretching of the vessels, they activate efferent parasympathetic nerves and deactivate efferent sympathetic nerves, which leads to a loss of total peripheral resistance due to vasodilatation. Also, it leads to a decrease in heartbeat frequency.

Long-term blood pressure regulation is mostly done by antidiuretic hormone (ADH), atrial natriuretic peptide (ANP), and the renin angiotensin aldosterone system (RAAS).

ADH is released by the posterior pituitary in states of hypernatremia or hypovolaemia. Chemoreceptors, which are in the hypothalamus, can recognise changes in the natrium threshold of 2 mOsm/L, and in the case of a hypernatremia they trigger ADH release and thirst. (4,5)

Baroreceptors in the carotid arteries, the aortic arch, and in the atrium can induce ADH secretion through the vagus nerve when a hypovolaemic state is noticed, as well as this they can inhibit ADH release when the body is hypervolemic.

After its secretion ADH is transported to vasopressin receptor 2 (V2) in the collecting duct cells, thus resulting in aquaporin 2 channels becoming phosphorylated and translocated to the apical plasma membrane. Those channels are making the membrane permeable to water, which makes a passive transport from water along the sodium chloride (NaCl) and urea gradient possible. When an isotonic state is restored, the ADH levels sink again, resulting in the channels moving into the cell again. (6)

Another way in which ADH regulates the blood pressure is via smooth muscle cells (SMCs). It binds to V1 receptors causing smooth muscle contraction. This results in a direct increase of blood pressure through vasoconstriction, but also secondarily through decreasing the renal blood flow and with it decreasing the amount of urine production. (4,5)

In pregnancy ADH secretion is elevated, but plasma levels usually stay normal due to an enzyme produced by the placenta, which is processing ADH, called vasopressinase. It was also noted that the osmoreceptors for ADH secretion and for thirst react more sensibly to changes in pregnancy. Serum levels of natrium are decreased by 4-5mEq/L and plasma osmolality lowers to around 270 mOsm/kg. (7,8)

ANP is produced and stored in cardiac myocytes, mostly in the right atrium, but also in the left and a very small amount in the cells of the heart chambers. Atrial stretch is one of the most important triggers for its release. (9,10)

The main effect of ANP consists in the relaxation of smooth muscle cells (SMC) of the arterioles, which together give an increase in the glomerular filtration rate (GFR). Furthermore, it induces natriuresis and diuresis and can inhibit the RAAS. (11,12) With those effects ANP is reducing the blood volume. Those effects alone would significantly increase the concentration of plasma proteins, which would eventually lead to an increase of vascular fluid through reabsorbing fluid form the interstitial space. To counter that, ANP increases the permeability of vessels, which helps plasma proteins go into the interstitial space, and with them vascular fluid, resulting in a decrease of blood volume as well. (13)

In pregnancy ANP was found to be elevated through stimulation by hypervolemia. Those changes were significant while the women were in a supine position, with no difference in upright posture. Also, that elevation lowered during pregnancy, due to decreased blood flow in the supine position because of the foetus compressing the vena cava. (14)

The RAAS System starts its activation with the enzyme renin, which is mostly produced and stored in the juxtaglomerular cell. It is released when the main arterial pressure in the kidney arteries decreases by more than 10-15 mmHg. Also, stimulation of β1-adrenoceptors through catecholamines leads to renin secretion as well as prostaglandin E (PGE), an important factor for renal blood flow. Renin transforms angiotensinogen, which is produced by the liver, into angiotensin I. Angiotensin I is transformed into Angiotensin II by angiotensinogen converting enzyme (ACE), which is mostly found in the lung tissue. Angiotensin II binds to angiotensin 1 (AT-1) receptors and induces potent vasoconstriction via increasing the intracellular calcium. This way Angiotensin II regulates short-term blood pressure adjustment, as well as this it stimulates the ADH and Aldosterone secretion for a long-term blood pressure adjustment. (15,16) Angiotensin II is the key effector role in the RAAS. (17)

Aldosterone is produced by the adrenal glands, in the zona glomerulosa. After its release it is mainly acting on the late distal tube of the nephron, as well as the collecting tube. It activates mineral corticoid receptors which lead to an increased expression of sodium channels, for an easier reabsorption of sodium, and with it water. For this passive transport of sodium, a concentration gradient is needed. This gradient is produced by sodium-potassium ATPase, which is responsible for transporting sodium into the vessels and potassium into the cells. This potassium can now be used to exchange sodium into the lumen of the kidney through potassium channels, eventually resulting in an increase of sodium and decrease of potassium in the body. (18) Here is a quick overview of the RAAS System as shown in Figure 2.

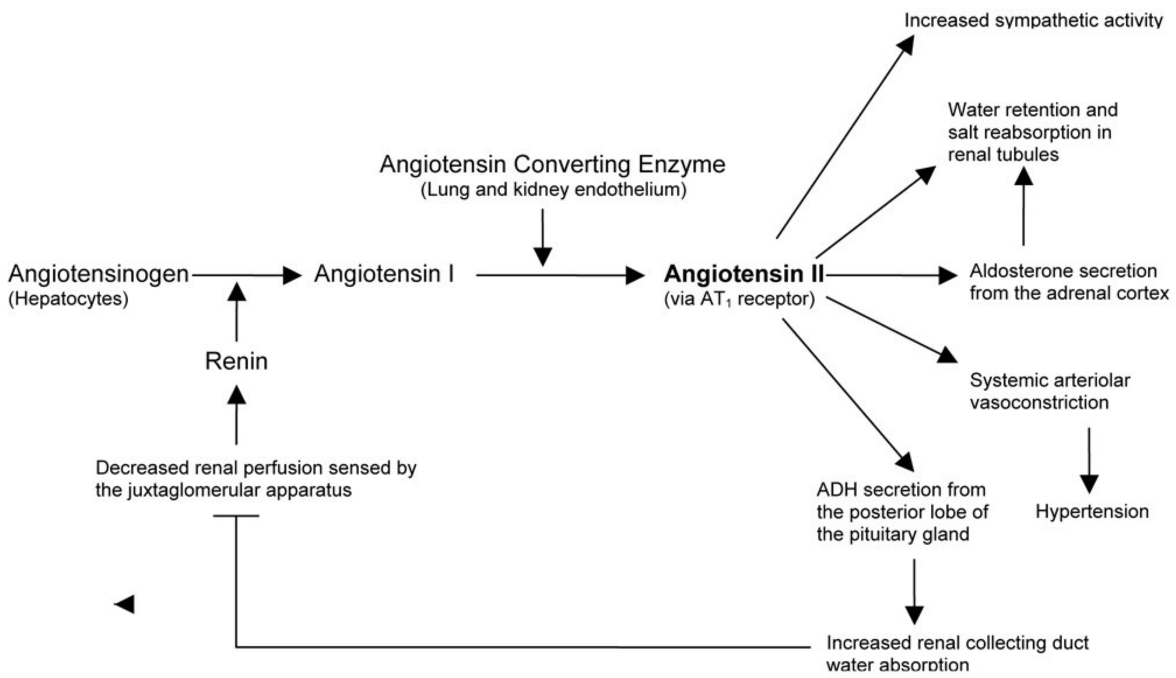


Figure 2: Overview of the RAAS

(Reproduced from: Irani, Roxanna A., und Yang Xia. The Functional Role of the Renin-Angiotensin System in Pregnancy and Preeclampsia. Placenta 29, Nr. 9 (September 2008): 763–71. https://doi.org/10.1016/j.placenta.2008.06.011.)

Pregnancy also has an impact on the RAAS. By now it is known that the components of this system are not exclusively produced in the locations mentioned before, but also in different tissues such as the ovary and the placenta. There was renin, prorenin and renin mRNA found in placental tissue, which indicates production from the placental tissue itself. (19) Furthermore angiotensinogen, which is usually produced in the Liver, angiotensin I, and angiotensin II have also been identified in placental tissue, as well as many of the involved receptors like AT1, renin, ACE, and angiotensinogen receptors. Because of this extra renal release, renin levels tend to increase early in pregnancy. As estrogen levels rise, the liver produces more angiotensinogen which then leads to an increase in angiotensin I and II, and aldosterone. The reason why the blood pressure is not rising in this case, but is usually lower in uncomplicated pregnancies, is partially due to a resistance to angiotensin II in pregnant woman. (20,21)

Angiotensin II also plays an important role in the development of PE. In this disease its serum levels are usually lower, but the body's response to it is more sensitive.

In conclusion the serum levels of ADH stay the same in pregnancy, while it is processed more and more because of vasopressinase. ANP is elevated during pregnancy in the supine position, while in an upright posture it is comparable with postpartum levels. Most components of the RAAS System are elevated in pregnancy, expect for ACE levels. There is a detailed review available published by Roxanna A. Irani and Yang Xia, which provides an overview of all the components and changes described in PE. (22)

5.1.1.3. Hematologic Adaptation

The increased blood flow is caused by an increase in plasma volume production of 45%, while red blood cell mass is increased by 30%, which leads all in all to an increase of approximately 1500 to 1600 ml in total blood volume. (23) The difference between these two results in the physiological anaemia of pregnancy. (24) Also pregnancy induces a state of hypercoagulability, resulting in an 3- to 4-times elevated risk of acute myocardial infarction. (25) This is discussed to be a physiological process that lowers the risk of perinatal bleedings. The state of hypercoagulability is due to different adaptations during pregnancies. On the one hand the concentrations of procoagulant factors are increased. This is described in factors V, VII, VIII, IX, X and XII as well as von willebrand factor. Also, fibrinogen levels are increased significantly in pregnancy. (26)

Anticoagulant factors are also affected by pregnancy, as their activity is decreased by it. (27,28) The euglobulin lysis time is a test which is used to represent the overall fibrinolysis activity of the body. This test needs more time in pregnancies, indicating the fibrinolytic activity is decreased, but it goes back to a normal range 30 minutes after delivery. (29,30) Furthermore, the prothrombotic state is affected by increased venous stasis due to compression of the vena cava and pelvic veins.

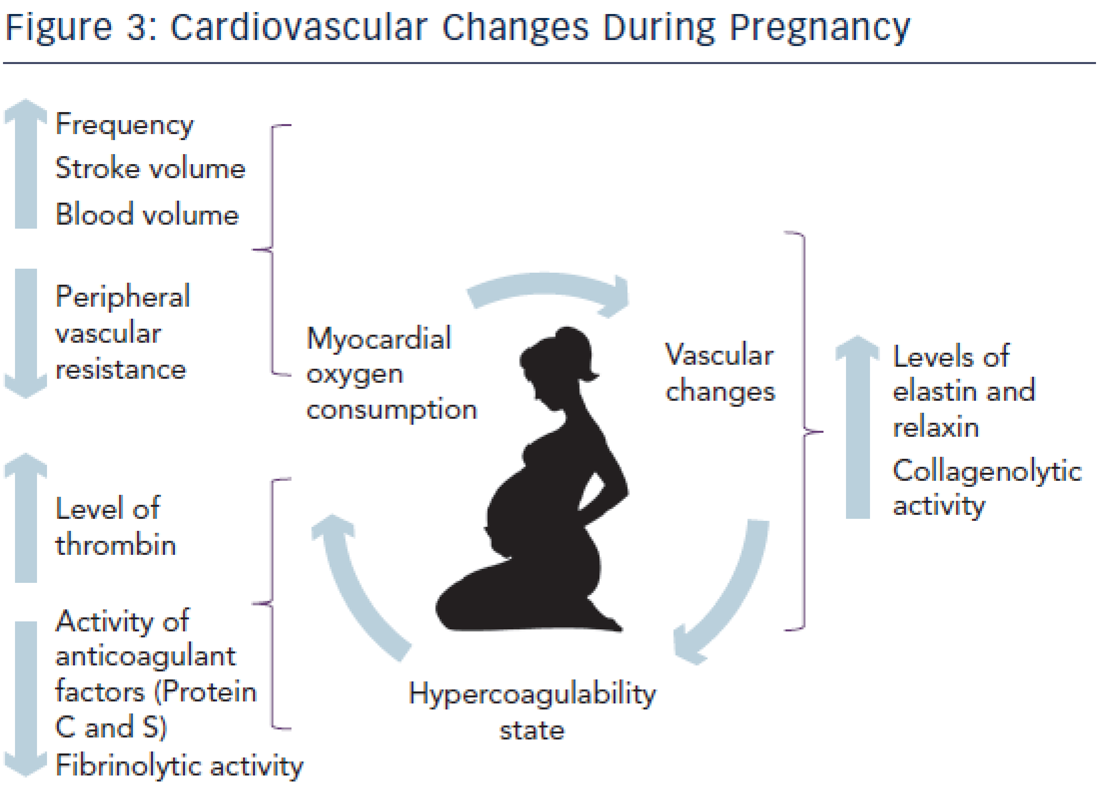


Figure 3:Cardiovascular Changes During Pregnancy

(Reproduced from: https://www.radcliffecardiology.com/image-gallery/figure-3-cardiovascular-changes-during-pregnancy (Access 14.03.2022))

5.1.1.4. Renal Adaptation

During pregnancy a woman undergoes a lot of adaptations, which are heavily influenced by the state of hypervolaemia and vasodilatation, which also explains some of the renal adaptations that occur in this state. Renal blood flow increases up to 80% as compared to non-pregnant woman, and the filtration rate increases by up to 50%. This leads to lower levels of serum creatinine, urea, and uric acid. More sodium is reabsorbed, which leads to a retention of 1.1 to 1.6 litres of water. Because of an increased vascular and interstitial volume, the length of the kidneys can expand by 1 to 1.5 cm during pregnancy. (8) It was thought that the growth of the kidneys is simply due to hydronephrosis in pregnant woman, but it was shown that also women without pelviectasis had an increased kidney size. (31)

The sodium retention is mostly due to an increased activity of the RAAS. The resorption of some substances is decreased, like glucose, amino acids, and some proteins, which can lead to an increase in urinary protein excretion of 150-180 mg/24h. That is why proteinuria in a pregnant woman is tolerated up to 300mg/24h without being pathological. Also, glucosuria can appear during pregnancy because of these changes. (32)

Anatomically the kidneys can retain 200-300 ml more urine because of the dilated collecting system. The range of the incidence rate of physiologic hydronephrosis is between 60-100% according to different studies, and the dilatations can cause stasis of urine, and thus result in an elevated risk of pyelonephritis. (8,33)

Proteinuria in pregnancy is especially important when it comes to PE, as it is one of the main symptoms. While it is considered to be normal when up to 300mg/d in a regular pregnancy, with a mean of about 150-180mg/d, in PE it is above 300mg/d.

And the reason for the higher levels of proteinuria in normal pregnancy might be similar to the causes of PE. Soluble fms-like tyrosine kinase-1 (sFLT-1) is produced by the placenta, which lowers the effects of vascular endothelial growth factor (VEGF). While VEGF shows a negative correlation to urinary albumin excretion, sFLT-1 shows the opposite, as with its rise also albumin excretion rises. (34) Because of their importance in the pathogenesis of PE, these two factors will be described later in more detail.

5.1.1.5. Hormonal Adaptation

Hormonal adaptations play an important role in many physiological adaptations during pregnancy. The female body starts preparing itself for the pregnancy with endocrine changes through the menstrual cycle and is dependent on those throughout the entire pregnancy. Those changes are responsible for many processes, like ensuring correct blood flow, the growing of the foetus, and management of the energy sources.

One of the first changes in the endocrine system is the production of **human chorionic gonadotropin** (hCG) by the developing embryo. This hormone prevents the decay of the corpus luteum, which can keep on producing progesterone and estrogen until the placenta can produce those hormones itself. (35)

Progesterone rises at the second half of the menstrual cycle, the luteal phase, to prepare the female body for the pregnancy. The corpus luteum is producing the progesterone in the first 10 weeks. This hormone is likely to be responsible for preparing the endometrium for an implantation and starting the process of decidualization. There were progesterone receptors found on the endothelium of decidual cells, and the binding to those promotes endothelial cell proliferation. (36)

Decidualization describes the transformation that endometrial stromal cells undergo to adapt for pregnancy and was described in a study undertaken with biopsies from endometrial cells in 1950 by Noyes, Hertig and Rock. It starts with edema in the superficial stroma in the first days of the luteal phase, and an increase of cytoplasm near the terminal spiral arteries. (37,38) This process results in an increase of permeability. The amount of natural killer (NK) cells also increases through progesterone. This also promotes the vascular development of the decidua because the uterine NK cells secrete several angiogenic factors such as VEGF-C, placental growth factor (PlGF), Angiopoietin-1 (Ang-1), Ang-2 and transforming growth factor β 1 (TGFβ1). (39)

Progesterone is an important vasodilator that regulates the vascular tone in the uterus during the first weeks of pregnancy. It induces nitric oxide (NO) synthase activity and production of NO in the endothelium, resulting in an increase of NO and therefore vasodilatation. Prostacyclin (PGI2) production is induced due to an upregulation of COX-1 and COX-2 activity with the same outcome. Prostaglandin E2 (PGE2) production in the myometrial tissue decreases. Furthermore, in the first trimester progesterone decreases the PGE2 production from the decidual stromal cells and inhibits the effects of Interleukin-1B (IL-1B) on COX-2 positive cells. In Vascular SMCs progesterone receptors were found inducing vasodilatation through a decrease of intercellular Ca2+. (36,40)

Estrogens are inducing angiotensinogen production by the liver. This leads to an elevation of aldosterone levels during pregnancy proportionally with the levels of estrogens. (23)

It is an important hormone for inducing placental angiogenesis and it is crucial for maintaining early pregnancy. The vascular resistance gets influenced by estrogen directly, but also by NO and PGI2 production which is induced by estrogens. When endothelial cells get exposed to estrogens, Nitric Oxide Synthase 3 (NOS3) and COX1 expression gets induced, which leads to a production of NO and PGI2. This causes vasodilatation and therefore a loss of vascular resistance. It is crucial for estrogen to promote angiogenesis in the beginning of the pregnancy and then switch to promoting vasodilatation, because this is the more important regulation factor of blood flow as gestation advances. (36)

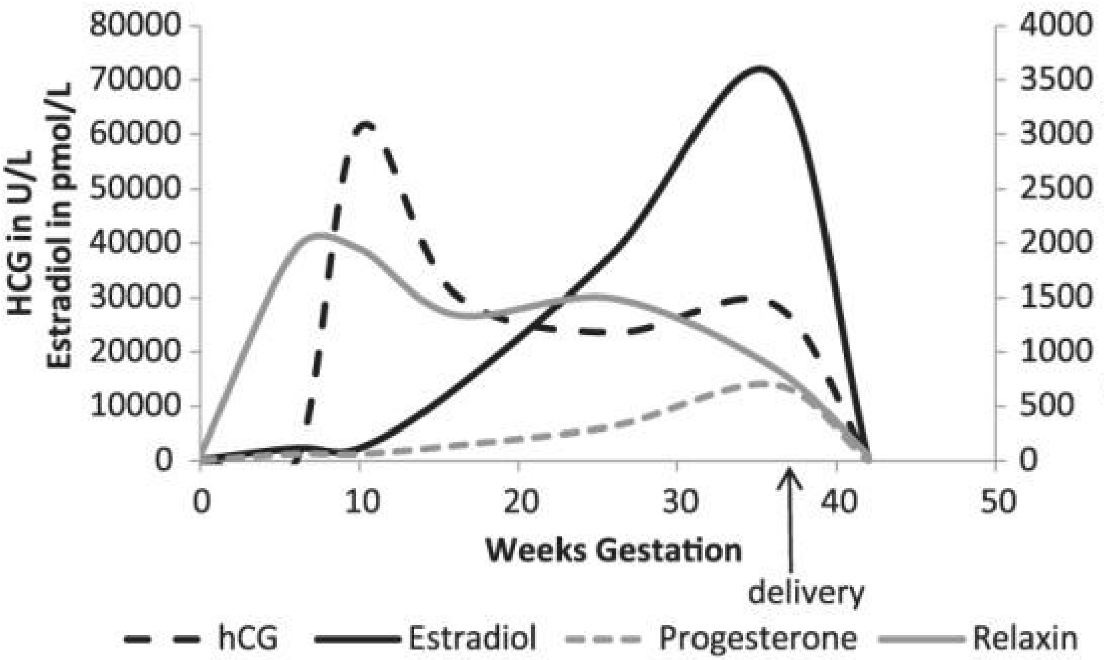


Figure 4: Hormone producion in pregnancy

(Reproduced from: Cheung, Katharine L., und Richard A. Lafayette. Renal Physiology of Pregnancy. Advances in chronic kidney disease 20, Nr. 3 (Mai 2013): 209–14. https://doi.org/10.1053/j.ackd.2013.01.012.)

5.1.2. Vascular Adaptation

In a pregnant woman the uterine blood flow ranges from 30- to 50-fold increases compared to a non-pregnant woman. This high blood flow is necessary to provide the foetus with sufficient nutrients and to facilitate the gas exchange. To reach those high levels of blood flow the body of a pregnant woman undergoes two crucial processes. One of them is angiogenesis, and with-it outward hypertrophy and lengthening of the vessels, and the other one is vasodilatation, which gets more important from mid gestation through parturition. (36,41)

5.1.2.1. Angiogenesis

Angiogenesis is the term used for the growth of vessels out of pre-existing vessels. This happens when the local balance between angiogenic factors and their inhibitors leans towards the pro-angiogenic side. Those angiogenic factors are common in tumor growth, and usually consist of cytokines and growth factors, but in a pregnant woman estrogens and hCG also play a role in angiogenesis. (36,41)

In pregnant women, more physiological changes of the uterine vessels come with angiogenesis. They undergo outward hypertrophy, which means an increase of the cross-sectional area while the wall thickness stays the same. That results in an increased lumen; seen for example in the main uterine arteries where the diameter doubles in pregnant women, as well as in the smaller arcuate and radial arteries where the lumen enlarges its diameter between 25 and 220%. (42) In a study by Stephen S. Ong et al. it was shown that preeclamptic women had small myometrial arteries with a reduced lumen diameter, a thicker wall, and a higher wall/lumen ration. (43)

One of the most important family of angiogenic factors is **VEGF**. (44) This family of proteins consists of VEGF-A, VEGF-B, VEGF-C, VEGF-D and the PlGF. The most important receptors of the family regarding angiogenesis are soluble fms-like tyrosine kinase-1: sFlt-1 (also called sVEGFR-1), and VEGFR-2. While the second one is responsible for the typical effects that VEGF is commonly known for, like augmenting permeability, anti-apoptotic and mitogenic effects on the endothelial cells, the first one is believed to lower the effects by binding the ligand without inducing the classic pro angiogenic effects of VEGF-A and PlGF, this is also known as decoy receptor. (41,44) Angiogenesis starts in the proliferative phase and continues in the secretory phase of the menstrual cycle, where VEGF levels are three times elevated compared to the proliferative phase. This elevation is partly due to progesterone and estrogen, which are inducing VEGF expression in human uterine stromal cells. (41,44) Other factors that induce VEGF expression are low oxygen levels, growth factors (e.g. TGF-α, TGF-β, IGF-1, FGF…) and cytokines, as well as hormones like the before mentioned progesterone and estrogen, but also hCG, TSH or ACTH. (45)

Zhang et al. showed that pregnancy can increase the effects of VEGF in vitro on human uterine artery endothelial cells as well as that it can augment the effects of the vasodilators NO and H_2S. (46)

Fibroblast growth factor is also a family of proteins which contains at least 18 different factors. Of which, FGF-1 and FGF-2 are known as the most important ones, stimulating the proliferation, the differentiation (both mostly on endothelial cells, but also muscle cells) and migration of cells, as they inhibit apoptosis. FBF has receptors responsible for inducing the function we know it for; FBFG 1-4, but also receptor types that mostly sequestrate and stabilize the ligand. (41)

The **angiopoietin** family consists of four members, Ang 1-4. Best known are Ang 1 and 2, and they work complementary to the VEGF system especially in the later stage of angiogenesis. In the pregnancy they are mostly produced by the placenta, and they have two regarding tyrosine kinase receptors called Tie-1 and Tie-2. Ang-1 has a stabilizing effect on the endothelium as it stimulates the reorganization of endothelial cells, but also influences and recruits peri-endothelial cells. Ang-2 is believed to act as a counterpart to Ang-1 as its main effect is to disconnect endothelium and perivascular cells, to induce apoptosis and to degenerate vascular structures. (47)

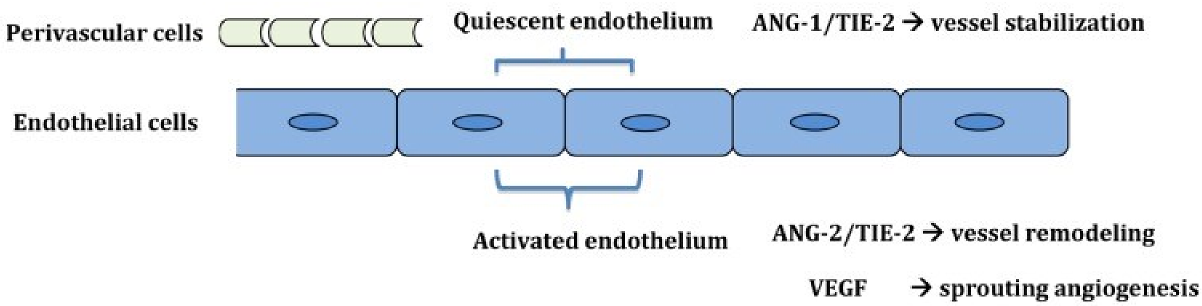


Figure 5: Role of the Angiopoietin/Tie System in Pregancy

(Reproduced from: D. KAPPOU u. a., Role of the angiopoietin/Tie system in pregnancy (Review),Experimental and Therapeutic Medicine 9, Nr. 4 (April 2015): 1091–96, https://doi.org/10.3892/etm.2015.2280.)

5.1.2.2. Vasodilatation

The circulation of organs is dependent on the arterial pressure and the vascular resistance of the organ. The vascular resistance is locally regulated by metabolic, chemical, and mechanical factors, which is especially important for blood flow in the placenta and uterus. Centrally the circulation is regulated via nerval and hormonal regulation with vasodilatation or construction. In the following, you can find a quick overview on the most important chemically working vasodilation factors. (48)

NO, formerly known as endothelium-derived relaxing factor (EDRF), is released through mechanical stress of the endothelial cells or directly through substances like bradykinin or prostaglandins binding to receptors. These stimuli activate Ca2+ channels, and free Ca2+ activates the release of NOS3. This induces that NO is made from arginine, O2 and NADPH by oxidation. As a soluble gas it diffuses then from the endothelium to the smooth muscle cell and induces vasodilatation via regulating the vascular smooth muscle tonus through cyclic guanosine monophosphate (cGMP) production. (48)

The activation of NOS3 plays a key role in releasing NO. This works with phosphorylation on multiple positions as well as the said high Ca2+ levels, to eventually produce NO. Phosphorylation alone is not sufficient, but in some activation sites, it can increase the sensitivity, as in S1179 and S617. Those positions are both described to be phosphorylated during pregnancy in uterine artery endothelial cells. (49)

Through COX-1 prostaglandin H2 is made from arachidonic acid, and via PGI2 synthase **PGI2** is made from prostaglandin H2. This metabolite reacts with prostacyclin receptors, resulting in smooth muscle relaxation. Thromboxane A2 (TXA2) works as its counterpart as it is inducing vasoconstriction. Prostacyclin, as well as COX-1 is both reported to be upregulated in pregnancies, showing that this pathway of vasodilatation plays a role in pregnant women. Also, the PGI2/TXA2 ratio is high, which results in vasodilatation as well. One way of inducing PGI2 production is via cytosolic phospholipase A2 (cPLA2) activation. The activation of this enzyme is dependent on high Ca^{2+} levels just like the activation of NO, which shows the importance of Ca^{2+} signaling in vasodilatation.

Endotheluim-derived hyperpolarizing factor (EDHF) is a pathway that leads to vasodilatation, and especially in pregnancy it plays a main role in providing enough blood flow to the uterus and the foetus. Despite its name, not a single factor is responsible for this hyperpolarization, but numerous endothelium-derived factures can induce this hyperpolarization, including NO and prostacyclin. As the other pathways, EDHF is also triggered by high Ca^{2+} levels, which activate potassium channels and end in the hyperpolarization of the endothelial cells, which causes them to relax and expand their diameter. In some blood vessels, but not in all, it is also able to hyperpolarize the underlying smooth muscle cells, which means that it is dependent on the vascular bed or size. (50)

5.1.3. Placental Development

Approximately the fifth day post fertilisation (dpf) the trophectoderm (TE) forms, which is the outer layer of the blastocyst. This structure is where the human placenta will develop from. When it attaches to the endometrial tissue, one part of the TE will differentiate into syncytiotrophoblast cells, invading the maternal decidual stroma and forming the primary syncytium. On approximately the 14^{th} dpf the blastocyst is entirely covered by surface epithelium. While making its way into the decidua, decidual uterine spiral arteries get remodelled, forming fluid and blood-filled spaces in the syncytial mass which are called lacunae and will form the intervillous space. (36,51)

The other cells of the TE, now lying beneath the syncytiotrophoblast are called cytotrophoblast cells. Those proliferate and push through the primary syncytium forming primary villi and giving the name to the next phase – the villous stage of development. This tissue will keep on forming more villous trees and proliferates to the outer layer of the primary syncytium to surround all the primary syncytium, thus building a cytotrophoblast shell between the built primary villi and the decidua. (51)

At around 17-18 dpf the secondary villi are made. This happens through extraembryonic mesenchymal cells, which lie under the cytotrophoblast, and start to grow into the primary villi. Those extraembryonic mesenchymal cells can now develop into blood vessels, forming the tertiary villi.

The transformation of the spiral arteries is very important considering the pathogenesis of preeclampsia. This process starts with smooth muscle swelling and endothelial vacuolation. Then, within the first 14 dpf cells start emerging from the cytotrophoblast shell and from the anchoring villi to invade the decidua. Those cells can be divided into interstitial and endovascular extravillous cells (iEVT and eEVT). The iEVT go to the maternal spiral arteries through the decidua, while the endovascular ones move inside the spiral arteries. The EVT are now preparing the spiral arteries with the goal to reach a continuous high blood flow with low pressure, to not damage any parts of the placenta. This is done by fibrinoid changes through a loss of actin in the smooth muscle cells in the arterial media, which leads to less activity in the vessel leaving it dilated. Because the full circulation of the foetus is not completely established at this time, eEVT cells migrate downstream of the modified artery to form a plug until around the 9th week of gestation. (51,52) After the 9th week the plug decays and the modified uterine spiral arteries supply the intervillous space with maternal blood. (38)

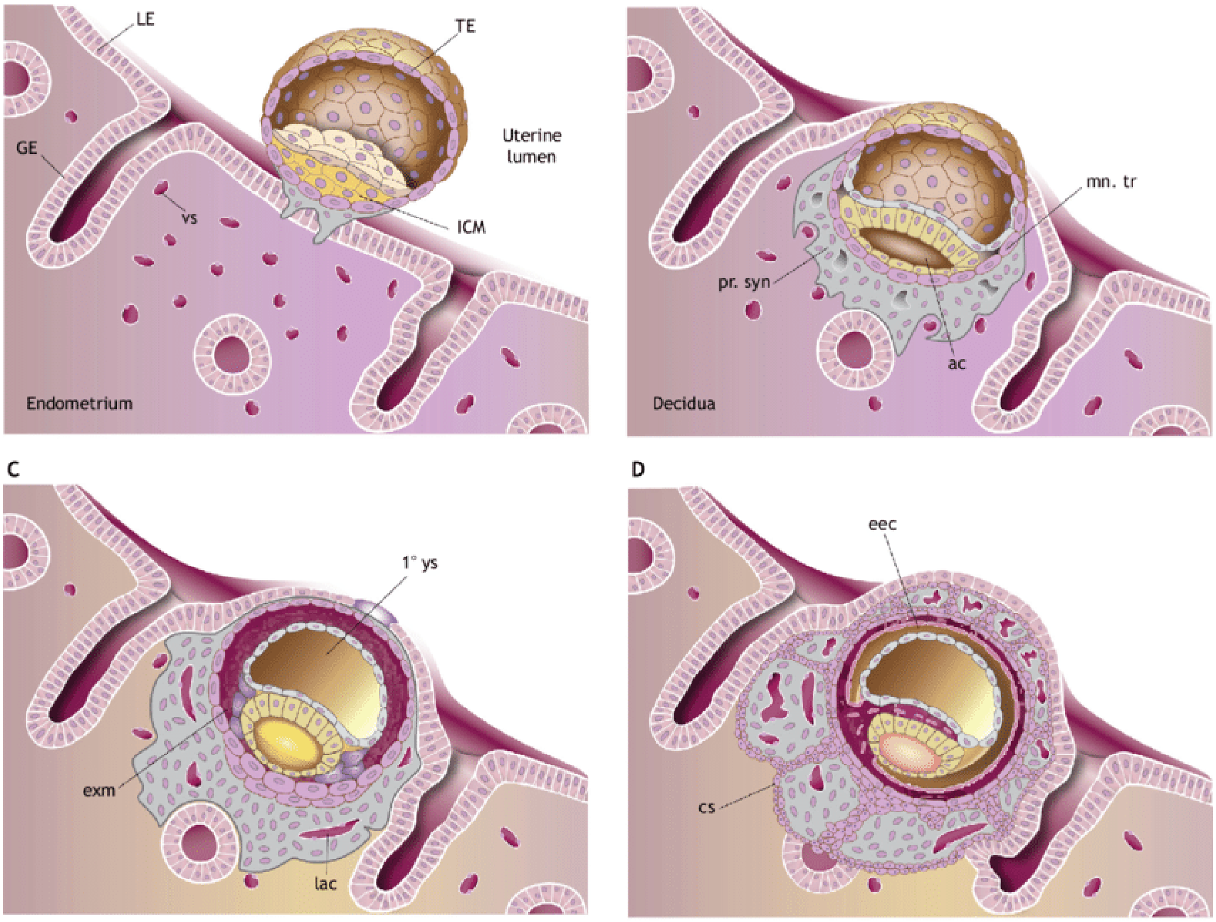


Figure 6: Early Development of the Placenta

(A,B) The pre-lacunar stages. (C) The lacunar stage. (D) The primary villous stage.
1° ys, primary yolk sac; ac, amniotic cavity; cs, cytotrophoblastic shell; eec, extra-embryonic coelom; exm, extra-embryonic mesoderm; GE, glandular epithelium; ICM, inner cell mass; lac, lacunae; LE, luminal epithelium; mn. tr, mononuclear trophoblast; pr. syn, primary syncytium; TE, trophectoderm; vs, blood vessels.

(Reproduced from: 1. Turco M, Moffett A. Development of the human placenta. Development. 2019 Nov 15;146:dev163428.)

5.2. Preeclampsia

Preeclampsia is a disease which is characterized by hypertension and proteinuria starting after the 20. gestational week in women who were normotensive and previously without proteinuria. (53)

5.2.1. Epidemiology

Many different risk factors have been identified for PE. Those named here are the most commons, as well as the most important ones regarding this thesis.

Maternal specific risk factors are pre-existing conditions of the mother, such as age. It was shown that Mothers older than 40 years are close to having double the risk of developing PE. Also, the parity is influencing the risk of suffering PE. Nulliparity women were shown to have close to triple the risk.

Previous PE comes with a 7-fold risk of developing PE, and having a family history of PE increases the risk by 3 times. (54)

Many of the classic risk factors for cardiovascular disease also raise the risk for PE, like pre-existing hypertension, pregestational diabetes (risk depending on severity), obesity, and renal disorders. Considering the impact of obesity there is a review study by Arum Jeybalan et al. that goes into depth about obesity as a risk factor of developing PE. (55,56)

Pregnancy associated risk factors can also influence the risk of PE, like the development of the placenta. Excess volume of the placenta is considered to raise the risk for suffering PE, like it is the case for multifetal gestation or hydatidiform moles. (56)

As exogenous factors stress and working status, socioeconomic status as well as smoking status are considered to increase the risk of suffering PE.
It was shown that women who worked during pregnancy were more likely to develop PE. Depression and anxiety are also associated with an increased risk of suffering PE. (57,58)

While the negative effects of smoking during a pregnancy are well known, it is associated with a decreased risk of suffering PE of around a third (32%), as a systematic review by Conde-Agudelo et al. demonstrated. (59) It is important to say that the smokers with PE also had higher rates of perinatal loss, abruptio placentae, low birth weight and small for gestational age, when compared to non-smoking women with PE. This shows just some of the adverse effects that smoking has on an unborn child.

5.2.2. Prevalence

This is the fourth biggest reason for maternal death, after thromboembolism, haemorrhages, and infections. The prevalence is associated with the country income. Overall, the prevalence of PE is 6.7% (95% CI=5.8–7.6) in high-income countries, while in a medium-income country the prevalence is 10.6% (95% CI = 6.05%–16.2%). Low-income countries have the highest prevalence with 11.5% (95% CI = 7.8%–15.8%) as shown in the review from Macedo et al. (60)

5.2.3. Classification

Hypertension in pregnancy can have three main forms:

Chronic hypertension: onset before pregnancy or diagnosed before 20 weeks of gestation.

Gestational hypertension: starting de novo after 20 weeks of gestation without proteinuria and without biochemical or haematological abnormalities, this can also be transient gestational hypertension.

Preeclampsia: de novo hypertension (>140/90) after 20 weeks of gestation or superimposed on chronic hypertension and either one of the following criteria or combinations (proteinuria is not mandatory for the diagnosis):

- Proteinuria of more than 300mg/day
- Maternal organ disfunctions, such as: Liver disfunction, acute kidney injury, neurological symptoms, haemolysis, or thrombocytopenia
- Uteroplacental disfunction such as fetal growth restriction

HELLP syndrome: This syndrome is considered as a severe form of preeclampsia and not as a separate disease. It consists of the symptoms: **h**aemolysis, **e**levated **l**iver enzymes, **l**ow **p**latelets. (53,61)

The more the pregnancy progresses, the higher the risk for women with preeclampsia to suffer **eclampsia**. This is the term for the onset of generalized seizures in women suffering preeclampsia, and it comes with a high mortality rate for the mother and the child.

Maternal mortality in developed countries range from 0-1,8%, while in developing countries the mortality for the mother reaches up to 15%. (62)

The seizures usually last a minute, they can be followed by more seizures and the women may fall into a coma after or between seizures. (63)

Preeclampsia has also been classified as being both severe and mild, but this classification is not recommended anymore because both forms can pose a big threat to the baby as well as to the mother, so there is a danger of misleading the patient. (61)

5.2.4. Pathogenesis

The pathogenesis of preeclampsia starts with a disturbed development of the placental vasculature resulting in hypoperfusion and ischaemic conditions of the placenta. The diseased placenta is now releasing anti-angiogenic factors into the maternal circulation which further leads to systemic endothelial disfunction and microangiopathy in the mother. (64,65)

The first step mentioned here of the pathogenesis is the disturbed development of the placental tissue. One hypothesis is that the trophoblastic invasion of the spiral arteries is not sufficient, and the changes are reduced to the decidua, while in normal pregnancy they extend to the myometrium. This can lead to hypoxia and ischemia in the placenta. This can also directly affect the unborn child and induce intrauterine growth restriction due to reduced placental perfusion. It is to point out that these effects are not sufficient to produce preeclampsia alone, as placental hypoxia does not have to trigger preeclampsia, and can be absent in some cases of preeclampsia. (65,66)

In response to stress (ischemia and hypoxia) the syncytiotrophoblast releases anti-angiogenic factors like soluble endoglin (sEng), sFlt-1, and inflammatory cytokines.

sFlt-1, as described above, works as a decoy receptor, meaning that it binds do VEGF-A and PlGF and inhibits its function by that way. It was seen that sFlt-1 was significantly higher and free PlGF was significantly lower in preeclamptic women compared to normotensive pregnancies. (58,67,68)

sEng functions as a negative regulator of TGF-β1. That means that the typical angiogenic effects of TGF-β1 are suppressed, leading to an anti-angiogenic effect, to a reduced activation of eNOS and less vasodilation. Furthermore, it was seen to induce permeability of the vessels and hypertension. (69)

sEng was seen to be up to four times higher in preeclamptic women and started to increase significantly 2 to 3 months before the onset of preeclampsia. In addition, higher levels of sEng showed higher ratios of sFlt1:PlGF. Especially, the combination of the two are predictors for the development of preeclampsia. (70)

There are many findings which indicate that the release of those anti-angiogenic and vasoconstrictive factors contribute to endothelial damage of the maternal vessels and produce the symptoms of preeclampsia, such as: hypertension, proteinuria, endothelial dysfunction and with it glomerular endotheliosis, and the injury of the kidney from preeclampsia. (66,71) One finding for example, was the appearance of these symptoms after exogenous application of sFlt-1 in rats done by Maynard et al.. (72)

The damage of the endothelium can be shown by serum markers which are higher in endothelial damage or activation like von willebrand antigen, cellular fibronectin, soluble tissue factor, soluble E-selectin, platelet derived growth factor, and endothelin, all of which are higher in preeclamptic women. (71)

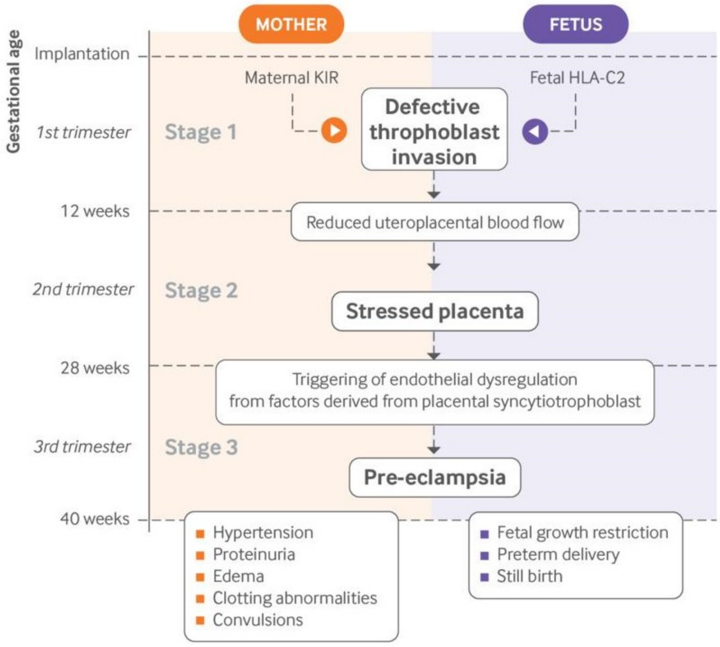


Figure 7: Pathogenesis of Preeclampsia

(Reproduced from: Burton GJ, Redman CW, Roberts JM, Moffett A. Pre-eclampsia: pathophysiology and clinical implications. BMJ. 2019 Jul 15;366:l2381.)

Due to high levels of sFlt-1, the kidneys also suffer damage which can be seen as glomerular endotheliosis and podocyte injury, resulting in proteinuria. Glomerular endotheliosis consists of swollen endothelial of the glomeruli and mesangial cells, as well as obstructed capillary lumens and a reduction in fenestration of endothelial cells. Podocyte injury is the second factor contributing to proteinuria. Podocytes are working as a sieve in the glomerulus and regulate the permeability of the glomerulus. They produce podocyte specific VEGF, which the effect of is impaired in preeclamptic women due to sFlt-1. Because of this, endothelial cells get damaged and release endothlin-1, leading to podocyte injury, podocyturia, and proteinuria. (73)

The hypertension in preeclampsia is due to high sFlt-1 levels, endothelin-1 release, reduced NO levels and many other factors, but not the RAAS-system, as renin, aldosterone and angiotensin levels are lower in preeclamptic women compared to normal pregnancy. (67) Still, there are some differences regarding this system. Angiotensin II type 1 receptor agonistic antibodies (AT1-AAs) were found to be elevated in women with preeclampsia, as well as women with abnormal uterine perfusion. These antibodies can activate the AT1 receptors making them more sensitive to AT2 and subsequently increasing the downstream effects of it. (74) The AT1-AAs form preeclamptic women or RUPP (reduced uterine perfusion pressure) rats could induce the preeclampsia phenotype in rats, which showed reduced GFR, more oxidative stress, the elevation of angiogenic factors like sFlt-1 and sEng, an increase in blood pressure, and reduced pup weight. (75)

5.2.5. Diagnosis and Symptoms

There are first trimester screenings of preeclampsia, consisting of an evaluation of maternal risk factors, blood pressure, PlGF and pregnancy-associated plasma protein A (PAPP-A) levels, and uterine artery doppler, for evaluating which persons will benefit from 150mg/day of aspirin to prevent pre-term preeclampsia. (61)

Hypertension in pregnancy is diagnosed by measuring blood pressure. This, together with urine stripe tests, is the main screening recommendation for preeclampsia in the 2^{nd} and 3^{rd} trimester. For the diagnosis, the blood pressure must be elevated on repeated occasions, and it is recommended to confirm the diagnose with 24h blood pressure measuring to exclude white coat hypertension. Also, the guidelines point out to choose the right size of the sphygmomanometer and to measure both arms.

If there’s more than 1+ protein detected in the dipstick urinalysis test then the amount of protein should be quantified with either protein-creatinine ratio or quantified in 24h-hour urine collection. If proteinuria ≥ 300mg/d is detected, no further quantification is necessary because it has no predictive value to the disease.

Oedema is a symptom of value for the diagnosis of preeclampsia if it is growing fast (>1kg/week in the third trimester) or if the face is heavily affected.

Furthermore, a lot of laboratory parameters can be affected due to preeclampsia. Kidney parameters like creatinine or uric acid can be elevated, liver damage can be shown by AST, ALT or bilirubin elevation, and haemoglobin and haematocrit can be elevated due to the hypovolemia seen in preeclamptic women. (76)

5.2.6. Therapy

There are preventive, antihypertensive, and anticonvulsive therapy options for preeclampsia.

For preventing preeclampsia in women who are at high risk of developing this disease, they get a low dose of aspirin (usually 150mg/d). Especially when one of the following conditions is recognized: previous pre-eclampisia, antiphospholipid antibody syndrome, multiple pregnancy, obesity, pre-existing maternal conditions like renal disease or chronic hypertension. Aspirin should be administered before the 16th week of pregnancy. Furthermore, these women are recommended to exercise 3 days a week as doing so showed a trend towards risk reduction. (61)

When the blood pressure rises over 160/110 mmHg, antihypertensive therapy should be started immediately. There is no clear consensus if mild to moderate hypertonia (140-159/90-109 mmHg) should be treated, the Austrian, German, and Swiss societies for gynaecology and obstetrics recommend a target blood pressure of 130-150 mmHg systolic and 80-100 mmHg diastolic. (76)

Anticonvulsive therapy consists of intravenous MgSO4 application which was shown to approximately half the rate of eclampsia and is recommended as first line therapy for seizure prevention. (61)

5.3. Smoking

Smoking can have different forms. The most common use of smoking tobacco products worldwide is cigarette smoking. Smoking cigarettes is so common due to its easy access all over the world and its addictive nature. Cigarettes make deep inhalations possible and can flood the brain with nicotine in 10 to 20 seconds, making it the most addictive way of consuming tobacco. (77)

It is well known that cigarette smoking causes serious harm for a human body. It can cause cardiovascular and pulmonary disease, harmful effects on the reproductive system, numerous types of cancer, and many more illnesses.

5.3.1. Disease

Regarding the cardiovascular system smoking cigarettes can induce many diseases, including: coronary heart disease, hypertension, strokes, aneurysms, or peripheral arterial disease. These diseases are due to the various effects that smoking has on our body. One of which is inducing endothelial dysfunction. Via oxidative stress, nicotine, and free radicals, the availability of nitric oxide is reduced. This leads to a less responsive endothelium to stress factors and therefore to a disturbed vasodilation or vasoconstriction. Furthermore, lead, arsenic, and mercury, among other ingredients of cigarette smoke, can lead to cellular damage and endothelial dysfunction. Smoking also has a prothrombotic effect. This is due to the activation of the coagulation system and the activation and aggregation of platelets. Lipid oxidation, insulin resistance, and a proinflammatory state induced by smoking are also effects contributing to the cardiovascular diseases developing through inhaling cigarette smoke. (77,78)

The respiratory tract is also affected by smoking cigarettes. Smoking is the main risk factor for suffering chronic obstructive pulmonary disease. It is also associated with suffering asthma and tuberculosis. (79,80) This relies on several mechanisms. Smoking tobacco is known to reduce the number of cilia in the lungs, leading to impaired clearance of the lungs. Also, it leads to mucus gland hyperplasia, chronic airway inflammation, increased production of IgE, and bronchial hyperresponsiveness. (77)

The DNA is also affected by smoking. This can lead to increased cell growth while suppressing the mechanisms which should inhibit cell growth and spread, leading to cancer. While it is well established that the main risk factor for lung cancer is smoking, the evidence is sufficient that smoking cessation can decrease the risk of several other cancer types like laryngeal cancer, cancer of the oral cavity and pharynx, esophageal, pancreatic, bladder, stomach, colorectal, liver, cervical, and kidney cancer as well as the risk of suffering acute myeloid leukemia. (78)

5.3.2. Smoking in Pregnancy

The main pathophysiological factors causing disease in pregnant mothers who smoke are carbon monoxide, tar, and nicotine.

While smoking a cigarette carbon monoxide can cause baseline carboxyhaemoglobin levels of up to 3-8%, while non-smokers usually have an average of 1%. This shifts the oxygen-haemoglobin dissociation curve to the left, leading to a decreased oxygen delivery to the myometrium and therefore to the foetus, which can lead to fetal growth restrictions or preterm birth. (81)

Tar, besides damaging the respiratory tract of the mother, was shown to be fetotoxic and teratogenic in animal studies and can accumulate in the placenta impairing the growth of the foetus and thus leading to growth restriction.

Nicotine can affect the placental vasculature, the fetus, and can also cause alterations in the brain and lung development, leading to cognitive, behavioural, and emotional problems as well as impaired lung development. (81,82)

In 1957, one of the first studies on smoking and pregnancies showed that smoking mothers give birth to babies with a reduced birth weight. (83,84) Many other studies confirmed this afterwards and showed the other effects that smoking can cause to an unborn child. By now the cessation of smoking is recommended generally, and especially when pregnant. It was shown that smoking has a significant effect on the birth weight of the foetus resulting in a lower birth weight (LBW), as the birth weight decreases as the number of daily cigarettes increases. This study shows that mothers who smoke 6-10 cigarettes a day bear babies with a reduced mean birth weight of 320 g, while it was 435 g when smoking 11-40 cigarettes a day. (85)

Also, the risk of giving birth to a baby that is small for the gestational age (SGA), which means to have a birth weight below the 10th percentile gestational age, has an odds ratio of 2.41. (86,87)

The chances for preterm birth are higher when the mother is a smoker, and the risk for placenta previa, abruptio placenta, ectopic pregnancy and preterm premature rupture of the membranes increases. The risk of perinatal death, stillbirth and neonatal death is also increased due to smoking (87,88)

Lung damage as well was reported due to maternal smoking, likely because of nicotine passing the placenta and interacting with nicotinic acetylcholine receptors in the fatal lung. This can prohibit the foetus to develop the full potential of its lungs which predisposes it to pulmonary disease. (82)

Also, behaviour of the children is affected by smoking, as it was seen in a study by batstra et al. children of smoking mothers during pregnancy showed more signs of attention deficit, higher levels of troublesome behaviour and performed worse at arithmetic and spelling tasks. (89)

6. Aims and Objectives

Smoking is a serious harm for the mother and the child, still, it was shown to have a protective effect on preeclampsia, a disease which can be life threatening for the pregnant mother and her offspring. While every pregnant mother should be advised to stop smoking during their pregnancy, as well as before and afterwards, it is important to question the pathways of the protective effect of smoking on preeclampsia. This can be helpful for understanding the pathophysiological processes involved in this disease, which still have many questions to answer, but also to find possible therapy options.

This thesis is a systematic review of the literature on the topic “the effects of smoking on preeclampsia” from the last 10 years. To structure the following part better it will be divided into four different sections. First, the information from cohort studies about smoking and preeclampsia of the last 10 years will be reviewed. It will be commented on as to whether the risk was elevated or decreased, and it will point out any possible interfering factors such as the age of the mother, obesity, or pre-conception smoking exposure. This part aims to review the risk reduction of preeclampsia.

The second part will be about angiogenic, or inflammatory factors found to be altered due to smoking in the placental tissue or the serum of pregnant women as well as in animal studies. This part has the objective to review the found literature on the mechanisms of smoking, nicotine and carbon monoxide altering the angiogenic balance or inducing inflammatory states in the tissues and to help understand pathophysiological processes in preeclampsia.

Afterwards, two papers concerning racial differences in the prevalence of preeclampsia and the impact of smoking on it will be discussed. Even though the National Institutes of Health Revitalization Act in 1993 was made to include more women and minorities in studies, often the study populations are still not representative and include less minorities than white men. (90) Therefore, those articles will be reviewed and explained if any racial differences are found.

And lastly, two articles referring to a possible left truncation bias in the studies already done on preeclampsia and smoking will be discussed. This is about a systematic bias in the previous studies which could have led to a false understanding of the effects of smoking on the risk of suffering preeclampsia.

7. Methods

This thesis is a systemic review of the available literature on the effects of smoking on preeclampsia. For the introduction, books and e-books were provided by the library of the Medical University of Graz, publications found via PubMed, Google Scholar and Web of Science, information was supplied by the World Health Organization, guidelines, and literature from the International Society for the Study of Hypertension in Pregnancy and from the Austria, Swiss and German society of Gynecology and Obstetrics were used.

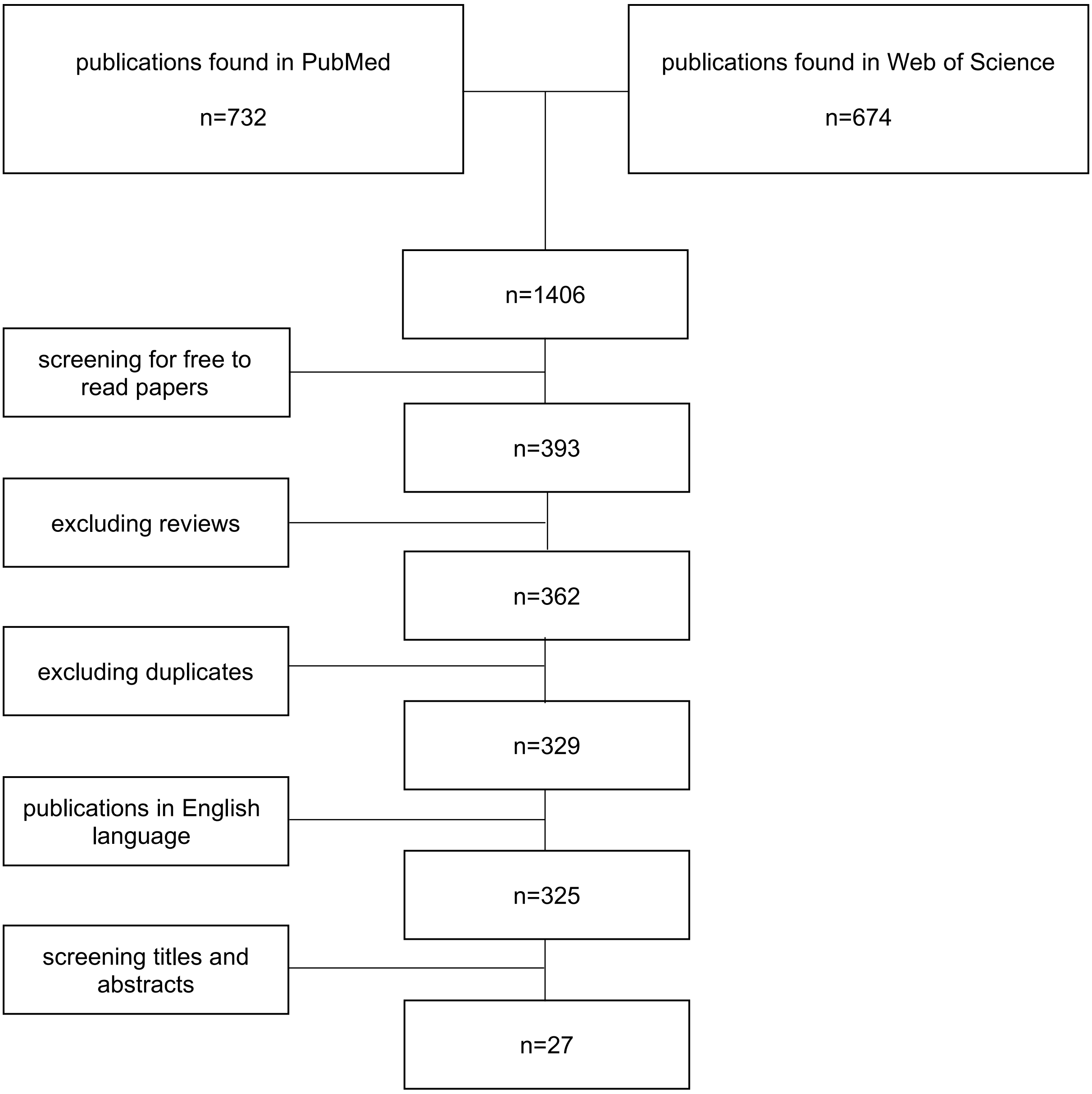
To put emphasis on the more recent findings the available literature of the last 10 years was systemically reviewed using a free text search in PubMed and Web of Science containing the words “smoking preeclampsia”. In this first approach 674 PubMed and 674 Web of Science articles were found.

Additionally, the following MeSh terms while searching the PubMed database were used: “Smoking”, “Tobacco Smoking”, “Nicotine”, “Carbon Monoxide”, “Preeclampsia”, “Eclampsia” combining them with the operators “AND” and “OR”. This brought up 58 results. Articles regarding nicotine and carbon monoxide were included since they are important factors regarding the effects of cigarette smoke.

After this search process an overwhelming number of 1406 articles published between 2011 and 2021 were found. To narrow down the number of found publications the following criteria was applied.

- Freely available articles
- Publications focusing on cigarette smoke, nicotine, and carbon monoxide were elected
- Articles available in English
- Publications regarding preeclampsia and eclampsia

The said criteria was applied via going through the titles and the abstracts, the duplicates were eliminated, resulting in a final number of 27 Publications. Additionally, I screened all the found publications for relevant secondary literature. If it was freely available and matched the criteria, the information was included in the discussion.


publications found in PubMed
n=732
publications found in Web of Science
n=674
n=1406
screening for free to read papers
n=393
excluding reviews
n=362
excluding duplicates
n=329
publications in English language
n=325
screening titles and abstracts
n=27

8. Review of Current Literature

8.1. Identifying the risk reduction of smoking in preeclampsia

Table 1: Identifying the risk reduction of smoking in preeclampsia

Author	Title	Study Type	Population	Location	Results
Perni et al. (2012)	Interpregnancy Change in Smoking Habits and Risk of Preeclampsia: A Population-Based Study.	retrospective population-based study	329,584	Sweden	Reduced OR to suffer preeclampsia for women who smoked**: OR: 0.54** (smoked in both preg.), **OR: 0.76** (smoked in sec. preg.), **OR: 0.81** (smoked in first preg.)
Gudnadóttir et al. (2016)	Body Mass Index, Smoking and Hypertensive Disorders during Pregnancy: A Population Based Case Control Study.	retrospective population-based case-control study	483 cases 962 controls	Iceland	Obese (BMI >30) women who smoked had a higher OR for suffering Preeclampsia than non-smokers. **OR 2.19** (obese non-smoking women), **OR 3.41** (obese smoking women) **Results statistically not relevant (p-value: 0.077)**
Fikadu et al. (2020)	Family history of chronic illness, preterm gestational age and smoking exposure before pregnancy increases the probability of preeclampsia in Omo district in southern Ethiopia: a case-control study.	retrospective case-control study	167 cases 352 controls	Ethopia	Smoking before pregnancy increased the risk **(OR: 4.19)** of suffering preeclampsia compared to women who never smoked. **Sample size among active smokers n=11**

Kharkova et al. (2017)	First-trimester smoking cessation in pregnancy did not increase the risk of preeclampsia/eclampsia: A Murmansk County Birth Registry study.	retrospective registry-based study	39 566	Murmansk (Russia)	Decreased OR for smokers: 1-5 cig/day= adjusted OR **0.69**, 6-10 c/d =**0.65**, ≥11c/d=**0.49** No significant difference between smoking during pregnancy and cessation in the first trimester
Ebbing et al. (2017)	Risk factors for recurrence of hypertensive disorders of pregnancy, a population-based cohort study	retrospective population-based cohort study	742 980	Norway	**No effect** on preterm preeclampsia in second pregnancy. **Small protective effect on HDP** (GH+PE) in second pregnancy with no HDP, term, or late preterm PE in first pregnancy
Weinberg et al. (2017)	Season of Conception, Smoking, and Preeclampsia in Norway.	retrospective registry-based study	356,662	Norway	Smoking **decreased risk** of suffering PE compared to non-smokers Smokers: **OR 0.61** Quitters: **OR 0.91** Quitters compared to smokers: **OR 1.50**
Lewandow-ska and Więckows-ka (2020)	The Influence of Various Smoking Categories on The Risk of Gestational Hypertension and Pre-Eclampsia.	prospective cohort study	cases: 137 controls: 775	Poland	Smokers: OR 0.91 (95%CI: 0.29–2.88), Ex-Smokers: OR 0.31 (0.04–2.40) Quitters during pregnancy: OR 2.25 (0.2–24.66) All the findings regarding preeclampsia are **statistically not relevant**

Ananth et al. (2013)	Pre-eclampsia rates in the United States, 1980-2010: age-period-cohort analysis.	retrospective population-based study	120 million births	USA	**Decrease in smoking** is associated with a **rise in preeclampsia** between 1987 and 2010
Lamminpää at al. (2013)	Smoking among older childbearing women - a marker of risky health behaviour a registry-based study in Finland.	retrospective registry-based study	11 277 > 35 years 68 983 < 35 years	Finland	Women <35 years: OR for smokers vs. non-smokers: **0.73**, 0.70-0.76 Women ≥35 years: OR for smokers vs. non-smokers: **0.83**, 0.66-0.93

In 1975 the Swedish Medical Birth Register was established. Perni et al. used it to analyze 371627 women who successfully gave birth to their first and second child between 1992 and 2006. Its goal was to see if a change in the smoking status can affect the risk of preeclampsia in two consecutive pregnancies. The here given OR was adjusted for covariates. The OR for women to suffer preeclampsia when they were smoking in both pregnancies was 0.54 (95% CI 0.47- 063), for women who smoked in the first pregnancy it was 0.81 (0.70-0.94) and for women who smoked in the second 0.76 (0.58-0.99). This supports the theory that the inverse association between preeclampsia and smoking is casual. (91) Gudnadóttir et al. did a study in 2016 finding contrary results to the study of Perni et al. regarding obese women. The study was carried out to evaluate if the risk reduction of smoking regarding preeclampsia is the same in obese women as in normal weight women. The results showed that the risk of getting preeclampsia increases with the body mass index, which was an already known risk factor for preeclampsia. But it also showed that smoking, instead of lowering the risk, might increase the risk of suffering preeclampsia in overweight and obese women. It is important to mention, that these findings were not statistically significant (p value=0.077), which means that this interaction still needs to be studied more before drawing conclusions. (92) To find out how smoking before conception modifies the risk of preeclampsia Fikadu et al. did a case-control study on women in southern Ethiopia. This study showed that smoking before conception increases the risk of suffering preeclampsia. Women who smoked before pregnancy had an adjusted OR of 4.16 to suffer preeclampsia with a p-value of 0.03 compared to women with no history of smoking. There were 519 women included in the study, of the eleven active smokers before pregnancy seven were suffering preeclampsia later and four were not. This low sample size should be considered when interpreting the results of this study. (93) Kharkova et al. found contrary results in a study made in 2017. However, here they looked especially at the effects of smoking cessation in the first trimester, and not at a general history of smoking before pregnancy like Fikadu et al.. In this study women were grouped into Non-Smokers, Quitters before Pregnancy (first trimester cessation) and Smokers. The odds for suffering preeclampsia/eclampsia for smokers and quitters were lower than non-smokers, and there was no significant difference found between the two groups. That means that the cessation of smoking in the first trimester did not increase the risk of suffering preeclampsia/eclampsia in this study compared to women who kept on smoking throughout their pregnancy. (94) Like in the study by Perni et at., investigating how smoking affects the risk of suffering preeclampsia in a second pregnancy was part of a study done by Ebbing et al. in 2016. Here, women were put into categories due to conditions regarding their first pregnancy, the groups were the following: No HDP, GH, term PE, late term PE, and preterm PE. There was no significant risk lowering effect of smoking for suffering preterm preeclampsia in a second pregnancy. The only effect of smoking was observed on term HPD, which includes GH and term PE, if the women suffered no HDP (OR 0.8 (95%CI 0.76–0.91)), term (0.8 (OR 0.67–0.96)), or late term PE (OR 0.6 (0.41–0.98)) in a first pregnancy. This categorization makes it hard to draw conclusions regarding preeclampsia, since it puts GH and term PE in one category. (95) Weinberg et al. investigated if the season of conception interferes with the risk of suffering preeclampsia. To make sure the findings are reliable and not affected by smoking, parity, fetal sex and longitude (region) they intended to replicate the pattern across these subpopulations. While doing so their results were similar to those of the study by Perni et al.. The overall estimated relative risk in smokers throughout the pregnancy was 0.61 (95% CI: 0.57, 0.65). Quitters appeared to have a higher risk than non-smokers, but it is mentioned in the study that this might be confounded due to a higher rate of having their first pregnancy in this group. The estimated adjusted risk for quitters was 0.91 (95% CI: 0.85, 0.97) compared to non-smokers, and 1.50 (95% CI: 1.38, 1.63) compared to smokers. In the next table an overview of the OR of quitters compared to non-smokers and smokers is given, however it is important to mention that the quitters in the study of Kharkova et al. quit smoking in the first semester, while in the studies of Fikadu et al. and Weinberg et al. quitter means history of smoking, and the differences might be due to the time when smoking was stopped. (96)

Table 2: Quitters vs. non-smokers

	Quitters vs. non-smokers	Quitters vs. smokers
Kharkova et al.		OR 1.10 (95%CI: 0.91–1.32)
Perni et al.	OR 0.81 (95%CI: (0.70, 0.94)	
Fikadu et al.	OR 4.16, (95%CI: 1.1–15.4)	
Weinberg et al.	OR 0.91 (95% CI: 0.85, 0.97)	OR 1.50 (95% CI: 1.38, 1.63)

Lewandowska and Więckowska did a study in 2020 using a Polish cohort of pregnant women to see how smoking affects the risk of preeclampsia. They divided the women into different groups: women who have never smoked, smokers (all smokers before and during pregnancy), ex-smokers (women who quit smoking before pregnancy), smokers in the first trimester, those who quit smoking during pregnancy (in II-III trimester), and smokers who reduced smoking during pregnancy (in II-III trimester). While this study design would be able to provide a lot of insight on how the cessation of smoking affects the risk of preeclampsia, due to the small sample size of preeclamptic women (n=24) all the found results are statistically not relevant

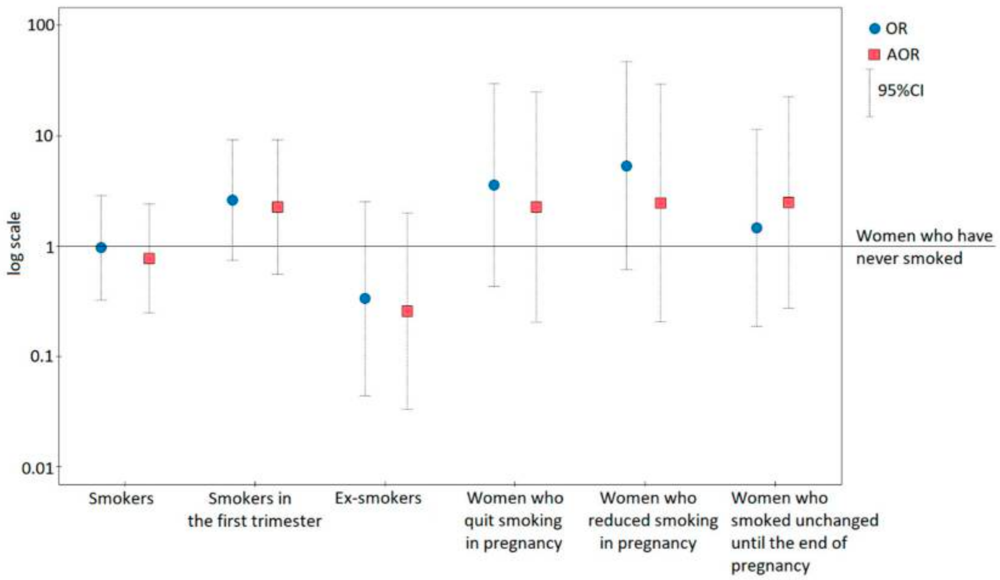


Figure 8: The Influence of Various Smoking Categories

(Reproduced From: Lewandowska M, Więckowska B. The Influence of Various Smoking Categories on The Risk of Gestational Hypertension and Pre-Eclampsia. J Clin Med. 4. Juni 2020;9(6):1743.)

P-values for the AOR for the categories: Smokers (0.872), Smokers in first trimester (0.208), Ex-smokers (0.260), Women who quit smoking in pregnancy (0.508), Women who reduced smoking in pregnancy (0.474), Women who smoked unchanged until the end of pregnancy (0.418). (97) An age-period-cohort study was carried out by Ananth et al. (2013) to see how the incidence of preeclampsia changed between 1980 and 2010. The preeclampsia rate increased from 3.4% to 3.8%, while the severe cases especially rose from 0.3% to 1.4%. In the time span between 1987 and 2010 the smoking level decreased from 30.3% to 18.7%, and the obesity prevalence rose from 21.7% to 36.0% between 1986 and 2010. Those factors alone are not sufficient enough to explain the rise in preeclampsia, but smoking was considered to increase the period effect for mild and severe cases of preeclampsia, which was noticed by a rise in preeclampsia cases across the different age categories, while the effects of obesity was seen more in younger categories due to a cohort effect, especially for severe preeclampsia. (98) Lamminpää at al. did a study in 2013 to find out how smoking affects the risk of preeclampsia over different age categories. The women were put into 4 groups, considering of non-smokers and smokers <35 and ≥35 years. In both smokers' groups the OR was decreased compared to the non-smokers. Women under 35 years who smoked had an AOR of 0.73 (95%CI: 0.70-0.76) compared to non-smokers, while women ≥ 35 years had an AOR of 0.83 (95% CI: 0.66-0.93). (99)

8.2. Pathophysiology

Table 3: Pathophysiology

Author	Title	Study Type	Population/ Sample Size	Location	Results
Jääskeläinen et al. (2017)	Angiogenic profile and smoking in the Finnish Genetics of Pre-Eclampsia Consortium (FINNPEC) cohort.	prospective study	trimesters 1st: 455 2nd/3rd: 228	Finland	Decreased sEng in smokers, decreased PlGF and **sFlt-1/PlGF** ratio in PE smokers
Tsiakkas et al. (2015)	Serum placental growth factor in the three trimesters of pregnancy: effects of maternal characteristics and medical history.	prospective study	trimesters 1st: 38 002 2nd: 10 281 3rd: 12 392	UK	**PlGF** usually lower in PE women, elevated in smoking mothers, especially in PE
Tsiakkas et al. (2015)	Serum soluble fms-like tyrosine kinase-1 in the three trimesters of pregnancy: effects of maternal characteristics and medical history.	prospective study	trimesters 1st: 7 066 2nd: 8 078 3rd: 10 464	UK	**sFlt-1** lowered in smoking mothers, especially in PE
Zhao et al. (2017)	Nicotine promotes vascular endothelial growth factor secretion by human trophoblast cells under hypoxic conditions and improves the proliferation and tube formation capacity of human umbilical endothelial cells.	cell-culture study			Low dose nicotine decreases **sFlt1** in human trophoblast cells under hypoxic conditions, promotes **VEGF** via induced mRNA transcription

Kahn et al. (2011)	Smoking in preeclamptic women is associated with higher birthweight for gestational age and lower soluble fms-like tyrosine kinase-1 levels: a nested case control study.	cell-culture study			Decreased sFlt-1 levels due to smoking
Zhao et al. (2021)	Maternal Immune System and State of Inflammation Dictate the Fate and Severity of Disease in Preeclampsia.	prospective study	PE: 60 control: 48	China	Placental tissue: increased levels of Flt1 and Eng in preeclampsia, **sFlt-1 and sEng significantly increased** in PE smokers
Venditti et al. (2014)	Chronic carbon monoxide inhalation during pregnancy augments uterine artery blood flow and uteroplacental vascular growth in mice.	animal study	46		Chronic CO exposure on mice: **doubled uterine artery blood flow** and **augmented uteroplacental vascular diameters** and branching. VEGF/FLT1 ratio increased
Venditti et al. (2014)	Carbon monoxide prevents hypertension and proteinuria in an adenovirus sFlt-1 preeclampsia-like mouse model.	animal study	58		**CO** could **prevent HT** and proteinuria in all mice, no effect on: litter size, fetal resorption numbers and fetal or placental weights
Linzke et al. (2014)	Carbon Monoxide Promotes Proliferation of Uterine Natural Killer Cells and remodelling of Spiral Arteries in Pregnant Hypertensive Heme	animal study	55		**CO improved SA remodelling** and **normalized blood pressure** in Hmox mutant mice – possibly because of uterine natural killer cells responsible for SA remodeling

	Oxygenase-1 Mutant Mice.				
Laule et al. (2017)	Effect of nicotine on placental ischemia-induced complement activation and hypertension in the rat.	animal study	31		**No** significant **decrease in BP** due to nicotine treatment
Kraus et al. (2014)	Cigarette smoke-induced placental adrenomedullin expression and trophoblast cell invasion.	placental tissue study	22		Trophoblast cell invasion **increased** in cells treated with **cigarette smoke extract** or **AM,** less invasion with AM-inhibitors. AM overexpression in placental tissue in smokers. Effect not reproducible with nicotine alone
Ghazavi Emma (2014)	The protein expression of the nicotinic acetylcholine receptors in the human placenta, and effects of cigarette smoking and pre-eclampsia.	placental tissue study	23		Protein expression (nAChR subunits) in placenta altered due to smoking
R. Chanda et al. (2019)	Impact of smoking on smooth muscle content of placental vessels in preeclampsia.	placental tissue study	4		Smoking **did not increase** the amount of collagen and smooth muscle cell
Wright et al. (2015)	Serum free β-human chorionic gonadotropin in the three trimesters of pregnancy: effects of maternal characteristics and medical history.	prospective study	trimesters 1st: 94 985 2nd: 7879 3rd: 8424	UK	**Lower β-hCG levels** due to smoking in preeclamptic mothers in the **second trimester**

8.2.1. Alterations in vascular endothelial growth factors

The first publications included in this review are regarding important factors of the VEGF family and their receptors. Jääskeläinen et al. (2017) used data from the FINNPEC (Finnish Genetics of Pre-eclampsia Consortium) study to find out whether maternal serum concentrations of sFlt-1, PlGF, sEng or the sFlt-1/PlGF ratio are altered in women who smoke in PE and non-PE women. First, they showed that smoking reduces sEng, and in PE it induces higher PIGF serum concentrations, which can be interpreted as smoking having its angiogenic effects due to altered angiogenic factors. In line with these findings is that smokers showed to have decreased sFlt-1/PlGF ratios in PE pregnancy. It is remarkable that smoking altered the angiogenic factors depending on if the woman is preeclamptic or not. They also put used these findings in relation to the number of cigarettes smoked. There they found an association between the number of cigarettes smoked and higher PlGF levels in PE Women. Also, the more cigarettes were smoked, the lower the sFlt-1/PlGF ratio was in PE and the higher it was in non-PE women. (100) Tsiakkas et al. (2015) found similar results regarding PlGF. This study showed, that PlGF is usually lower in women who develop PE, but elevated in smoking mothers, especially in smokers developing PE. They did a similar study in the same year on sFlt-1 and found out that it was lowered in PE smokers. (101,102) To reproduce the effects on sFlt-1 and VGEF in cell culture, Hongbo Zhao et al. (2017) studied the effects of nicotine and hypoxia on human trophoblast cells. The findings show that sFlt-1 was reduced due to nicotine in hypoxic cells, just as the study of Jääskeläinen et al. showed in pregnant women due to cigarette smoking. Furthermore, VGEF secretion and mRNA transcription was promoted due to nicotine in hypoxic cells, supporting the hypothesis that the alteration of angiogenic factors contributes to the protective effect, especially the down regulation of sFlt-1 and up regulation of VGEF. But also, those effects were not able to be reproduced under normoxic conditions, showing that nicotine might need the hypoxic conditions to have a protective effect on preeclampsia. (103) This could also be a possible explanation as to why smokeless tobacco has no protective effect on preeclampsia. (104) Khan et al. (2011) found similar results regarding sFlt-1 levels, as they were lower in smokers than non-smokers in preeclamptic women. Also, it was shown that the birth weight for gestational age (BWGA) z-score of children from preeclamptic women was increased by 0.57 if the mother was a smoker, which was contrary to the effects of smoking in the control group (BWGA z-score -0.29), which supports the hypothesis that smoking lowers the effects of preeclampsia, while it might have contrary effects on normal pregnancy. (105) The studies discussed in this chapter by now show possible pathways to how smoking could reduce the risk of suffering preeclampsia. Xin Zhao et al. (2021) investigated the inflammatory effects of smoking and how they might increase the severity of preterm preeclampsia (PPE) compared to preterm pregnancy. Placental tissue was collected right after cesarean cuts and showed increased Flt-1 and Eng gene expression in PPE compared to the control preterm group regardless of smoking history. In contrast to that sFlt-1 and sEng levels were significantly higher in the smoker group compared to the non-smokers – which is also contrary to the studies mentioned before, where sFlt-1 was lower in smoker groups. It is to mention, that this study investigated exclusively in preterm preeclampsia, and the sample size can be considered low with 18 smokers out of 60 in the PPE group (8/48 in the control group). (106)

8.2.2. CO and Nicotine

To reproduce the effects of smoking in mice, Venditti et al. (2014) conducted two studies using mice models. The first one was done in 2013, to show the effects of low dose ambient CO levels on the uterine blood flow in mice. It was shown that the diameter of the following vessels was increased in the chronic CO exposed group: uterine arteries, spiral arteries, canal branches and radial artery branches. Also, the maternal uterine artery blood flow was increased in the chronic CO group. These changes in angiogenesis might be due to a lowered Flt-1:VEGF ratio found in the group exposed to chronic and acute CO, which might show a way of how cigarettes are preventing preeclampsia. The second study was done in 2014 in preeclamptic like mice models that show hypertension and proteinuria. The symptoms were induced via an adenovirus that led to an overproduction of sFlt-1. CO therapy could prevent hypertension completely in the AdsFlt-1 group as shown in Figure 8, also proteinuria was significantly lower in the CO group in AdsFlt-1 mice.

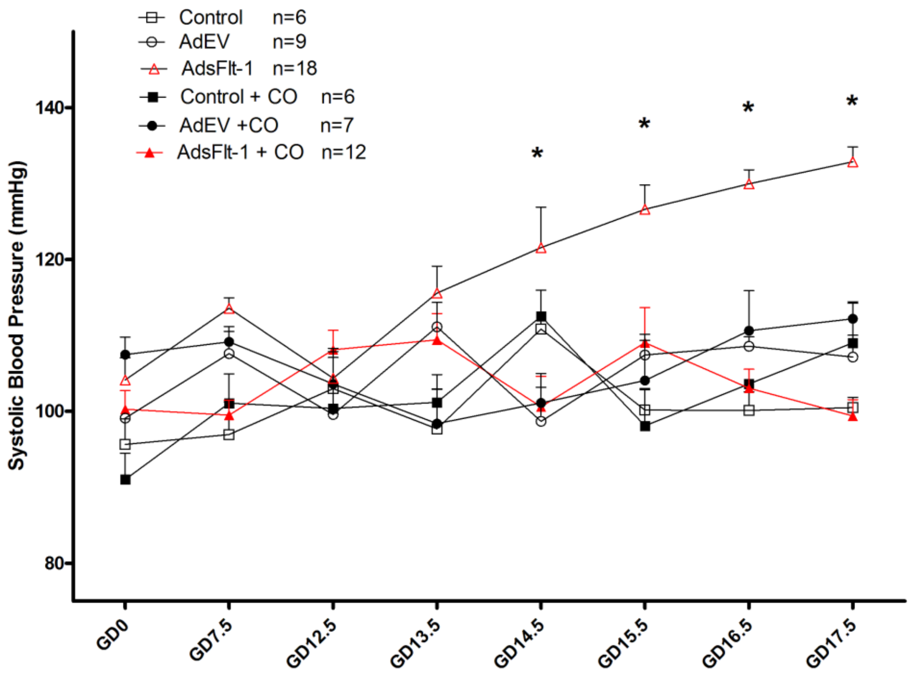


Figure 9: Effects CO Treatment in AdsFlt-1 Mice

(Reproduced From: Venditti CC, Casselman R, Young I, Karumanchi SA, Smith GN. Carbon monoxide prevents hypertension and proteinuria in an adenovirus sFlt-1 preeclampsia-like mouse model. PLoS One. 2014;9(9):e106502.)

It It is important to say that the litter sizes as well as the fetal/placental weight at birth was not affected by the CO treatment, and neither by the sFlt-1 Adenovirus, while studies on humans showed CO and PE can both lead to intrauterine growth restriction as previously mentioned in the introduction. These findings show that the effects of smoking could be induced by CO, which shows another pathway to the previously discussed study by Hongbo Zhao et al..

Another way to induce hypertension in mice is via impairing Heme-Oxygenase 1. (107,108) The study of Linzke et al. (2014) showed that in Heme-Oxygenase 1 knock-out mice CO treatment could avoid hypertension and promote normal fetal growth. Also, an increase in uterine NK cells was shown. Since those are responsible for SA modification (109), this might be a possible explanation for the protective effect of CO. Furthermore, SA modification was impaired in HO-1 knock knock-out mice. With this study, in two different mice models hypertension could be avoided due to CO exposure, supporting the hypothesis that CO plays an important role in the protective effect of smoking on preeclampsia. (110) In the study of Laule et al. in 2017, hypertension in rats was induced via reduced uteroplacental perfusion pressure (RUPP) and afterwards treated with subcutaneous nicotine or saline (control group) injections. In this study no statistical relevant difference was found between the nicotine and control group regarding BP, which shows again that CO might play an important role, and that nicotine might need hypoxic conditions to have protective effects on PE. (111)

Another pathway to the risk reduction of preeclampsia due to smoking could be Adrenomedullin, a potent vasodilator. Kraus et al. (2014) showed in a study that the trophoblast cell invasion is increased in cells treated with cigarette smoke extract or adrenomedullin, and that this invasion can be attenuated with AM-inhibitors. Also, it was shown that AM was overexpressed in the placental tissue of smokers. However, nicotine as a treatment alone could not increase trophoblast invasion, like in the studies mentioned before where the isolated application of nicotine did not have a preventive effect on preeclampsia. (112) Which effects nicotine can have on a human placenta depends on the receptors in the tissue, therefore Ghazavi Emma investigated in 2014 the different subunits of nicotinic acetylcholine receptors (nAChR) and the expressions in the human placenta. Receptor expression in normal, smoker, and PE placentas were compared. Compared to PE α2 and α9 expression was higher in smokers endothelial cells and/or villi, compared to control α7 expression which was higher in smokers and α9 was higher in PE in EC. Explaining those findings in depth would go beyond the scope of this discussion, but it is important to mention, that the differences found can result in different effects of nicotine in normal, hypoxic and preeclamptic mothers. If a receptor is more expressed in the placenta of PE mothers, nicotine could have stronger or attenuated effects, which might explain why some effects of smoking are exclusively seen in hypoxic or preeclamptic mothers. (113) Nicotine binding to nAChRs also induces smooth muscle cell proliferation in the airways, (114,115) while in preeclampsia vascular smooth muscle (VSMC) is decreased in placental tissue as R. Chada et al 2019 previously discovered. (95) To find out if smoking can reduce the risk of suffering preeclampsia due to inducing VSMC growth R. Chanda et al. investigated this topic. The result was that the decrease in VSMC was not attenuated due to nicotine exposure. (116)

8.2.3. β-hCG

Serum β-hCG levels were found to be elevated in preeclamptic mothers. (117) To find out what other factors can affect the β-hCG serum levels, and in which trimester they do so, Wright et al. (2015) did a study. It was shown that the β-hCG levels of smokers were lower in the first and second trimester. In women who developed PE the β-hCG levels were higher in the second and third semester. Smokers in the PE group had lower levels in the second semester, while the levels were higher in the third. (118)

8.3. Racial differences in smoking and preeclampsia

Table 4: Racial differences in smoking and preeclampsia

Author	Title	Study Type	Population	Location	Results
Chang et al. (2014)	Reassessing the impact of smoking on preeclampsia/eclampsia: are there age and racial differences?	retrospective cohort study	3,113,164 / 12,326,151	USA	OR only decreased in NH white and NH American Indian women under 35 years
Nakagawa et al (2016)	Racial/Ethnic Disparities in the Association Between Preeclampsia Risk Factors and Preeclampsia Among Women Residing in Hawaii.	retrospective cohort study	271,569	Hawaii	**OR 1.19** for smokers to suffer PE in Hawaiian residents

As mentioned before, minorities are not equally represented in clinical and biomedical research (90). To find out how this affects the studies conducted on smoking and preeclampsia Chang et al. did a study in 2014. It was shown that contrary to the belief that smoking reduces the risk of PE, this effect was just shown in non-hispanic white and non-hispanic American Indian women under 35 years of age as according to a study they carried out, the OR are shown in Table 5 below.

Table 5: Racial differences in smoking and preeclampsia

	Natality data (n = 3,113,164)		NIS data (n = 12,326,151)	
Women <35 yrs	**Adjusted OR***	**95 % CI**	**Adjusted OR⁺**	**95 % CI**
NH White	0.89	0.86, 0.91	0.95	0.91, 1.00
NH Black	1.05	1.00, 1.09	0.91	0.81, 1.03
NH American Indian	0.76	0.66, 0.87	0.96	0.64, 1.45
NH Asian/Pacific Islander	1.66	1.35, 2.05	1.36	0.96, 1.93
Hispanic	1.09	1.00, 1.20	0.99	0.84, 1.17
	Natality data (n = 3,113,164)		**NIS data (n = 12,326,151)**	
Women ≥35 yrs	**Adjusted OR***	**95 % CI**	**Adjusted OR⁺**	**95 % CI**
NH White	1.17	1.09, 1.26	1.29	1.13, 1.47
NH Black	1.18	1.01, 1.37	1.30	0.97, 1.62
NH American Indian	1.29	0.88, 1.89	1.07	0.25, 4.63
NH Asian/Pacific Islander	1.71	1.07, 2.72	2.32	1.35, 3.99
Hispanic	1.18	0.91, 1.54	0.91	0.57, 1.46

Abbreviation: OR, odds ratio, 95% CI, 95% confidence interval.
* adjusted for maternal age, marital status, parity, kotelchuck prenatal care index, gestational weight gain, chronic hypertension, diabetes.
⁺adjusted for maternal age, chronic hypertension, and diabetes.
doi:10.1371/journal.pone.0106446.t004

(Reproduced from: Chang JJ, Strauss JF, Deshazo JP, Rigby FB, Chelmow DP, Macones GA. Reassessing the impact of smoking on preeclampsia/eclampsia: are there age and racial differences? PLoS One. 2014;9(10):e106446.)

While these findings might show that many studies done on this subject could be biased by the study population, one limitation of the study is that Chang et al. did not differentiate between pregnancy induced hypertension and preeclampsia, which makes it harder to draw conclusions about preeclampsia. (119) Nakagawa et al. found similar results in 2016 as Chang et al., when considering the impact of race on the risk of developing preeclampsia in smokers. The study focuses on Hawaiian residents giving birth between the years of 1995 and 2013. There it was shown that across all of the study population, which was mostly Native Hawaiians (23.2%), but also Filipinos (19.5%), whites (18.9%), Japanese (11.4%), other Pacific Islander (8.2%), other Asian (5.1%), Chinese (3.4%) and other races (10.3%), smoking increased the risk of suffering preeclampsia by an OR of 1.19 (95%CI 1.07 – 1.33). Unfortunately, the study does not provide data on the single races but refers to the whole study population (all Hawaiian residents) when it comes to the effects of smoking, which makes it hard to interpret the data considering the different races involved. (120) While the data from those two studies suggests that race might play an important role regarding the effects of smoking, the discussed studies have important limitations, and there is still more investigation needed on this topic to draw conclusions.

8.4. Left truncation bias

Table 6: Left truncation bias

Author	Title	Study Type	Results
Lisonkova and Joseph (2015)	Left truncation bias as a potential explanation for the protective effect of smoking on preeclampsia. Epidemiology.	Simulation	Higher rates of early pregnancy loss result in a lower amount of preeclampsia in smokers in a simulation if 5 assumptions were applied
Kinlaw et al. (2017)	Left Truncation Bias to Explain the Protective Effect of Smoking on Preeclampsia: Potential, But How Plausible?	Simulation	If assumptions were slightly adapted RR changed greatly, showing that left truncation bias might play a less pronounced role

The last part of this thesis is about a possible left truncation bias in the studies on preeclampsia and smoking. Lisonkova and Joseph pointed this out in 2015, mentioning that a possible explanation for the risk reduction of preeclampsia in smokers is early pregnancy loss before the diagnosis of preeclampsia. To do so they performed two simulations with 5 underlying assumptions, one of which was that smoking does not affect abnormal placentation and therefore preeclampsia. The first simulation was based on data obtained from literature to estimate the risk of abnormal placentation and early pregnancy loss. This simulation resulted in consistent RR of PE in smokers of 0.85 (95% CI 0.73, 0.98). The second simulation was based on a ground rate of early pregnancy loss in non-smokers without abnormal placentation. Then the relative risks of abnormal placentation and smoking were applied. In the second simulation it was possible to show that higher rates of early pregnancy loss could result in a lower amount of preeclampsia in smokers, supporting the theory that this could have influenced studies before. (121) One important limitation of this study is the assumptions applied for the simulations, which were also mentioned by Kinlaw et al.

in 2017, when he responded to the paper by Lisonkova and Joseph. One of those assumptions was that abnormal placentation will develop preeclampsia and vice versa. If this risk of developing preeclampsia without abnormal placentation was increased from 0 to 1.02% the RR of preeclampsia in smokers increased from 0.84 to 0.92, showing that the assumptions that were made can have a big impact on the actual outcome. Also, Kinlaw et al. showed that the bias depends greatly on the baseline risk of early loss. As the RR for early loss in abnormal placentation rose from 1.5 to 2.0 to 2.5, the RR of suffering preeclampsia as a smoking mother changed from 0.96 to 0.92 to 0.88. In conclusion, this left truncation bias can still have an impact on the studies done on this topic, but Kinlaw et al. showed that it might not have the great impact as was shown by Lisonkova and Joseph. (122)

9. Conclusion

It is well known that smoking reduces the risk of suffering preeclampsia in pregnant mothers (123), as it was also shown in this review. Although one study showed that this effect does not apply on obese women, and another study showed that the effect was not seen in women who smoked before, but not during the pregnancy. (92,93)

How smoking reduces the risk of suffering preeclampsia is still not completely clear. This literature review showed that angiogenic factors might play an important role, as it was seen that smoking can alter the expression and concentrations of them. Except for one study from Zhao et al. in 2021 (106) where sFlt-1 was elevated in smokers, it was shown to be decreased due to smoking in preeclamptic mothers, while PlGF and VEGF were increased. (100–102,105) These effects might be partly due to nicotine, but to show the effects on cell cultures hypoxic conditions were needed, as demonstrated by Zhao et al. in 2017 (103).

CO seems to play a more important role than nicotine, considering the preventive effects on PE, as it could prevent HT and proteinuria, increase SA remodeling, and increase uterine blood flow in PE mice. Nicotine didn't show to have effects on BP under normoxic conditions, which supports the hypothesis that CO and hypoxic conditions play an important role in the risk reduction, and that nicotine might need hypoxic conditions to show effects on risk reduction. (107,108,110,111) This could be explained with different receptors expressed in the human placenta in smoking mothers. (113)

One aim of this study was to find out about racial differences in smoking and preeclampsia, because minorities are still underrepresented in clinical and biomedical research. (90) The studies reviewed in this thesis showed this as well. The effects of smoking when considering the risk of suffering preeclampsia or pregnancy induced hypertension was only seen in non-Hispanic white and non-Hispanic American Indian women under 35 years of age in a study done by Chang et al., and when the study population was not mostly white, there was even an increase seen in the OR from a study done by Nakagawa et al. (119,120).

Another topic discussed regarding this subject was the left truncation bias. Lisonkova and Joseph wanted to show, that the risk reduction of smoking might be due to early pregnancy loss. Even if this effect might play a role in the studies carried out on this topic, Kinlaw et al. showed that the underlying assumptions made in the simulation of

Lisonkova and Joseph had a big impact on the outcome of the ORs. Kinlaw et al. suggested that the underlying assumptions in the study are not that pronounced in real life, and therefore the simulation predicted a higher importance of the left truncation bias. (121,122)

References

1. Breckwoldt M, Schneider H. Schwangerschaftsveränderungen des mütterlichen Organismus. In: Breckwoldt M, editor. Gynäkologie und Geburtshilfe [Internet]. 2008 p. 5. Available from: https://doi.org/10.1055/b-001-2141.
2. Mahendru AA. A Longitudinal Study of Maternal Cardiovascular Function from Preconception to the Postpartum Period. J Hypertens. 2014 Apr;32(4):849-56,.
3. Grissmer S. Blutkreislauf. In: Behrends J, editor. Duale Reihe Physiologie [Internet]. 2017 p. 3 ,. Available from: https://doi.org/10.1055/b-004-132217.
4. Cuzzo B, Padala SA, Lappin SL. Physiology, Vasopressin [Internet]. Available from: http://www.ncbi.nlm.nih.gov/books/NBK526069/.
5. Davies AG. Antidiuretic and Growth Hormones. Br Med J. 1972 Apr 29;2(5808):282-84,.
6. Boone M, Deen PMT. Physiology and pathophysiology of the vasopressin-regulated renal water reabsorption. Pflugers Arch [Internet]. 2008;456(6). Available from: https://doi.org/10.1007/s00424-008-0498-1.
7. Davison JM. Serial Evaluation of Vasopressin Release and Thirst in Human Pregnancy. Role of Human Chorionic Gonadotrophin in the Osmoregulatory Changes of Gestation. J Clin Invest. 3(März 1988):798-806,.
8. Cheung KL, Lafayette RA. Renal Physiology of Pregnancy. Adv Chronic Kidney Dis. 3(Mai 2013):209-14,.
9. Kurtz A, Wagner C. Atriales natriuretisches Peptid (ANP). In: Behrends J, editor. Duale Reihe Physiologie [Internet]. 2017 p. 3 ,. Available from: https://doi.org/10.1055/b-004-132217.
10. Ruskoaho H. Release and Regulation of Atrial Natriuretic Peptide (ANP). Eur Heart J. 1987 May 2;8(suppl B):99-109,.
11. Cannone V. Atrial Natriuretic Peptide: A Molecular Target of Novel Therapeutic Approaches to Cardio-Metabolic Disease. Int J Mol Sci [Internet]. 2019 Jul 2;20(13). Available from: https://doi.org/10.3390/ijms20133265.
12. Ahmad F, Tabassum N, Rasool S. Regulation of atrial natriuretic peptide (ANP) and its role in blood pressure. Int Curr Pharm J. 2012 Jun 1;1:176-79,.

13. Curry F-RE. Atrial natriuretic peptide: an essential physiological regulator of transvascular fluid, protein transport, and plasma volume. J Clin Invest [Internet]. 2005 1;115(6). Available from: https://doi.org/10.1172/JCI25417.

14. Carla S. Atrial Natriuretic Peptide and Hemodynamic Changes During Normal Human Pregnancy. Hypertension. 1995 Apr 1;25(4):631-36,.

15. Horn F. Angiotensin II und das RAAS. In: Horn F, editor. Biochemie des Menschen [Internet]. 2018 p. 7 ,. Available from: https://doi.org/10.1055/b-006-160377.

16. Silbernagl S. Renin und Nierenhormone. In: Pape H-C, Kurtz A, Silbernagl S, editors. Physiologie [Internet]. 2019 p. 9 ,. Available from: https://doi.org/10.1055/b-006-163285.

17. Irani RA, Xia Y. The Functional Role of the Renin-Angiotensin System in Pregnancy and Preeclampsia. Placenta. 2008 Sep;29(9):763-71,.

18. Scott JH, Menouar MA, Dunn RJ. Physiology, Aldosterone. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2021. Available from: http://www.ncbi.nlm.nih.gov/books/NBK470339/

19. Hagemann A, Nielsen AH, Poulsen K. The Uteroplacental Renin-Angiotensin System: A Review. Exp Clin Endocrinol. 1994;102(3):252-61,.

20. Assali NS, Westersten A. Regional Flow-Pressure Relationship in Response to Angiotensin in the Intact Dog and Sheep. Circ Res. 1961 Jan;9:189-93,.

21. Gant NF. A Study of Angiotensin II Pressor Response throughout Primigravid Pregnancy. J Clin Invest. 1973 Nov 1;52(11):2682-89,.

22. Irani RA, Xia Y. Renin Angiotensin Signaling in Normal Pregnancy and Preeclampsia. Semin Nephrol. 1(Januar 2011):47-58,.

23. Soma-Pillay P. Physiological changes in pregnancy. Cardiovasc J Afr. 2016;27(2):89-94,.

24. Branch DW. Physiologic Adaptations of Pregnancy. Am J Reprod Immunol. 1992;28(3–4):120-22,.

25. H. JA. Acute Myocardial Infarction in Pregnancy. Circulation [Internet]. 2006 28;113(12). Available from: https://doi.org/10.1161/CIRCULATIONAHA.105.576751.

26. Paidas MJ, Hossain N. Hematologic Changes in Pregnancy. In: Hemostasis and Thrombosis in Obstetrics & Gynecology [Internet]. John Wiley & Sons, Ltd; 2010. p. 1-11,. Available from: https://doi.org/10.1002/9781444328332.ch1.

27. Oruç S. Changes in the Plasma Activities of Protein C and Protein S during Pregnancy. Aust N Z J Obstet Gynaecol. 2000 Nov;40(4):448-50,.

28. Aprigliano G. Acute Coronary Syndrome in Pre- and Post-Partum Women - A Review Interventional Cardiology Review 2013;8(1):8–13 Available from: https://www.icrjournal.com/articles/ACS-Pre-Post-Partum-Women.

29. Howie PW. Blood clotting and fibrinolysis in pregnancy.. Postgrad Med J. 1979 May;55(643):362–6.

30. Chang J, Streitman D. Physiologic Adaptations to Pregnancy. Neurol Clin. 2012 Aug;30(3):781-89,.

31. Christensen T. Changes in Renal Volume during Normal Pregnancy. Acta Obstet Gynecol Scand. 1989;68(6):541–3.

32. August P. Role of Renin-Angiotensin System in Blood Pressure Regulation in Pregnancy, Lancet [Internet]. Vol. 345. London, England; 1995. 896–97, p. Available from: https://doi.org/10.1016/s0140-6736(95)90012-8.

33. Peake SL, Roxburgh HB, Langlois SL. Ultrasonic Assessment of Hydronephrosis of Pregnancy.. Radiology [Internet]. 1983 Jan;1. Available from: https://doi.org/10.1148/radiology.146.1.6849041.

34. Yoshimatsu J. Relationship between Urinary Albumin and Serum Soluble Fms-like Tyrosine Kinase 1 (SFlt-1) in Normal Pregnancy. Eur J Obstet Gynecol Reprod Biol. 2006 Sep;128(1–2):204-8,.

35. Weissgerber TL, Wolfe LA. Physiological adaptation in early human pregnancy: adaptation to balance maternal-fetal demands. Appl Physiol Nutr Metab. 2006 Feb 1;31(1):1–11.

36. Boeldt DS, Bird IM. Vascular Adaptation in Pregnancy and Endothelial Dysfunction in Preeclampsia. J Endocrinol [Internet]. 232(1). Available from: https://doi.org/10.1530/JOE-16-0340.

37. Noyes RW, Hertig AT, Rock J. Dating the Endometrial Biopsy. Fertil Steril. 1950 Jan 1;1(1):3-25,.

38. Gellersen B, Brosens JJ. Cyclic Decidualization of the Human Endometrium in Reproductive Health and Failure. Endocr Rev. 6(1. Dezember 2014):851-905,.

39. Maliqueo M. Bárbara Echiburú, und Nicolás Crisosto, Sex Steroids Modulate Uterine-Placental Vasculature: Implications for Obstetrics and Neonatal Outcomes. Front Physiol [Internet]. 2016 Apr 26;7. Available from: https://doi.org/10.3389/fphys.2016.00152.

40. Noyola-Martínez N, Halhali A, Barrera D. Steroid Hormones and Pregnancy. Gynecol Endocrinol. 2019 May 4;35(5):376-84,.

41. Zygmunt M, Herr F, Münstedt K, Lang U, Liang OD. Angiogenesis and vasculogenesis in pregnancy. Eur J Obstet Gynecol Reprod Biol. 2003 Sep;110:S10–8.

42. Osol G, Mandala M. Maternal Uterine Vascular Remodeling During Pregnancy. Physiology. 2009 Feb;24(1):58–71.

43. Ong SS, Baker PN, Mayhew TM, Dunn WR. Remodeling of myometrial radial arteries in preeclampsia. Am J Obstet Gynecol. 2005 Feb;192(2):572–9.

44. Ylä-Herttuala S, Rissanen TT, Vajanto I, Hartikainen J. Vascular Endothelial Growth Factors. J Am Coll Cardiol. 2007 Mar;49(10):1015–26.

45. Ferrara N. Vascular Endothelial Growth Factor: Basic Science and Clinical Progress. Endocr Rev. 2004 Aug 1;25(4):581–611.

46. Zhang H, Chen JC, Sheibani L, Lechuga TJ, Chen D. Pregnancy Augments VEGF-Stimulated In Vitro Angiogenesis and Vasodilator (NO and H2S) Production in Human Uterine Artery Endothelial Cells. J Clin Endocrinol Metab. 2017 Jul 1;102(7):2382–93.

47. D. Kappou, S. Sifakis, A. Konstantinidou, N. Papantoniou, D. A. Spandidos. Role of the angiopoietin/Tie system in pregnancy (Review). Exp Ther Med. 2015 Apr;9(4):1091–6.

48. Behrends JC, Bischofberger J, Deutzmann R, Ehmke H, Frings S, Grissmer S, et al. Physiologie [Internet]. 3rd ed. Stuttgart: Georg Thieme Verlag; 2017 [cited 2021 Jul 9]. Available from: https://eref.thieme.de/10.1055/b-004-132217

49. Cale JM, Bird IM. Dissociation of Endothelial Nitric Oxide Synthase Phosphorylation and Activity in Uterine Artery Endothelial Cells. Am J Physiol-Heart Circ Physiol. 2006 Apr;290(4):1433-45,.

50. Félétou M, Vanhoutte PM. Endothelium-Derived Hyperpolarizing Factor. Arterioscler Thromb Vasc Biol. 2006 Jun 1;26(6):1215–25.

51. Turco MY, Moffett A. Development of the human placenta. Development [Internet]. 2019 Nov 27;146(22). Available from: https://doi.org/10.1242/dev.163428

52. Pijnenborg R, Vercruysse L, Hanssens M. The Uterine Spiral Arteries In Human Pregnancy: Facts and Controversies. Placenta. 2006 Sep;27(9–10):939–58.

53. Breckwoldt M, Schneider H. Schwangerschaftserkrankungen. In: Breckwoldt M, editor. Gynäkologie und Geburtshilfe [Internet]. 2008 p. 5. Available from: https://doi.org/10.1055/b-001-2141.

54. Duckitt K, Harrington D. Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies. BMJ. 2005 Mar 12;330(7491):565.

55. Sibai BM, Caritis S, Hauth J, Lindheimer M, VanDorsten JP, MacPherson C, et al. Risks of preeclampsia and adverse neonatal outcomes among women with pregestational diabetes mellitus. National Institute of Child Health and Human Development Network of Maternal-Fetal Medicine Units. Am J Obstet Gynecol. 2000 Feb;182(2):364–9.

56. Jeyabalan A. Epidemiology of preeclampsia: impact of obesity. Nutr Rev. 2013 Oct;71:S18–25.

57. Kurki T, Hiilesmaa V, Raitasalo R, Mattila H, Ylikorkala O. Depression and anxiety in early pregnancy and risk for preeclampsia. Obstet Gynecol. 2000 Apr;95(4):487–90.

58. Shamsi U, Saleem S, Nishter N. Epidemiology and risk factors of preeclampsia; an overview of observational studies. 2013;6:9.

59. Conde-Agudelo A, Althabe F, Belizán JM, Kafury-Goeta AC. Cigarette smoking during pregnancy and risk of preeclampsia: A systematic review. Am J Obstet Gynecol. 1999 Oct;181(4):1026–35.

60. Macedo TCC. Prevalence of Preeclampsia and Eclampsia in Adolescent Pregnancy: A Systematic Review and Meta-Analysis of 291,247 Adolescents Worldwide since 1969. Eur J Obstet Gynecol Reprod Biol. 2020 May 1;248:177-86,.

61. Brown MA, Magee LA, Kenny LC, Karumanchi SA, McCarthy FP, Saito S, et al. The hypertensive disorders of pregnancy: ISSHP classification, diagnosis & management recommendations for international practice. Pregnancy Hypertens. 2018 Jul;13:291–310.

62. Ghulmiyyah L, Sibai B. Maternal Mortality From Preeclampsia/Eclampsia. Semin Perinatol. 2012 Feb;36(1):56–9.

63. Cunningham FG, editor. Williams obstetrics. 24th edition. New York: McGraw-Hill Medical; 2014. 1358 p.

64. Al-Jameil N, Aziz Khan F, Fareed Khan M, Tabassum H. A Brief Overview of Preeclampsia. J Clin Med Res. 2014 Feb;6(1):1–7.

65. Sircar M, Thadhani R, Karumanchi SA. Pathogenesis of preeclampsia: Curr Opin Nephrol Hypertens. 2015 Mar;24(2):131–8.

66. Phipps E, Prasanna D, Brima W, Jim B. Preeclampsia: Updates in Pathogenesis, Definitions, and Guidelines. Clin J Am Soc Nephrol CJASN. 2016 Jun 6;11(6):1102–13.

67. Saleh L, Verdonk K, Visser W, van den Meiracker AH, Danser AHJ. The emerging role of endothelin-1 in the pathogenesis of pre-eclampsia. Ther Adv Cardiovasc Dis. 2016 Oct 1;10(5):282–93.

68. Levine RJ, Maynard SE, Qian C, Lim K-H, England LJ, Yu KF, et al. Circulating angiogenic factors and the risk of preeclampsia. N Engl J Med. 2004 Feb 12;350(7):672–83.

69. Liu Z, Afink GB, Dijke P ten. Soluble fms-like tyrosine kinase 1 and soluble endoglin are elevated circulating anti-angiogenic factors in pre-eclampsia. Pregnancy Hypertens Int J Womens Cardiovasc Health. 2012 Oct;2(4):358–67.

70. Levine RJ, Lam C, Qian C, Yu KF, Maynard SE, Sachs BP, et al. Soluble endoglin and other circulating antiangiogenic factors in preeclampsia. N Engl J Med. 2006 Sep 7;355(10):992–1005.

71. Young BC, Levine RJ, Karumanchi SA. Pathogenesis of Preeclampsia. Annu Rev Pathol Mech Dis. 2010 Jan;5(1):173–92.

72. Maynard SE, Min J-Y, Merchan J, Lim K-H, Li J, Mondal S, et al. Excess placental soluble fms-like tyrosine kinase 1 (sFlt1) may contribute to endothelial dysfunction, hypertension, and proteinuria in preeclampsia. J Clin Invest. 2003 Mar;111(5):649–58.

73. Moghaddas Sani H, Zununi Vahed S, Ardalan M. Preeclampsia: A close look at renal dysfunction. Biomed Pharmacother. 2019 Jan;109:408–16.

74. Campbell N, LaMarca B, Cunningham MW. The role of Agonistic Autoantibodies to the Angiotensin II Type 1 Receptor (AT1-AA) in Pathophysiology of Preeclampsia. Curr Pharm Biotechnol. 2018;19(10):781–5.

75. Cunningham MW, Castillo J, Ibrahim T, Cornelius DC, Campbell N, Amaral L, et al. AT1-AA (Angiotensin II Type 1 Receptor Agonistic Autoantibody) Blockade Prevents Preeclamptic Symptoms in Placental Ischemic Rats. Hypertension. 2018 May 1;71(5):886–93.

76. Gaß P. Hypertensive Schwangerschaftserkrankungen: Diagnostik und Therapie. :117.

77. Onor IO, Stirling DL, Williams SR, Bediako D, Borghol A, Harris MB, et al. Clinical Effects of Cigarette Smoking: Epidemiologic Impact and Review of Pharmacotherapy Options. Int J Environ Res Public Health. 2017 Sep 28;14(10):1147.

78. United States Surgeon General. The Health Consequences of Smoking -- 50 Years of progress: A Report of the Surgeon General: (510072014-001) [Internet]. American Psychological Association; 2014 [cited 2021 Oct 14]. Available from: http://doi.apa.org/get-pe-doi.cfm?doi=10.1037/e510072014-001

79. Wheaton AG, Liu Y, Croft JB, VanFrank B, Croxton TL, Punturieri A, et al. Chronic Obstructive Pulmonary Disease and Smoking Status — United States, 2017. MMWR Morb Mortal Wkly Rep. 2019 Jun 21;68(24):533–8.

80. Kim SY, Sim S, Choi HG. Active, passive, and electronic cigarette smoking is associated with asthma in adolescents. Sci Rep. 2017 Dec;7(1):17789.

81. McDonnell BP, Regan C. Smoking in pregnancy: pathophysiology of harm and current evidence for monitoring and cessation. Obstet Gynaecol. 2019;21(3):169–75.

82. McEvoy CT, Spindel ER. Pulmonary Effects of Maternal Smoking on the Fetus and Child: Effects on Lung Development, Respiratory Morbidities, and Life Long Lung Health. Paediatr Respir Rev. 2017 Jan;21:27–33.

83. Simpson WJ. A Preliminary Report on Cigarette Smoking and the Incidence of Prematurity. Am J Obstet Gynecol. 1957 Apr;73(4):807–15.

84. Floyd RL. A Review of Smoking in Pregnancy: Effects on Pregnancy Outcomes and Cessation Efforts. Annu Rev Public Health. 1993;14(1):379-411,.

85. Kataoka MC. Smoking during pregnancy and harm reduction in birth weight: a cross-sectional study. BMC Pregnancy Childbirth [Internet]. 2018 12;18(1). Available from: https://doi.org/10.1186/s12884-018-1694-4.

86. Mitchell EA. Smoking, Nicotine and Tar and Risk of Small for Gestational Age Babies. Acta Paediatr. 1992;91(3):323-28,.

87. Ko T-J. Parental Smoking During Pregnancy and Its Association with Low Birth Weight, Small for Gestational Age, and Preterm Birth Offspring: A Birth Cohort Study. Pediatr Neonatol [Internet]. 2014 1;55(1). Available from: https://doi.org/10.1016/j.pedneo.2013.05.005.

88. Pineles BL, Hsu S, Park E, Samet JM. Systematic Review and Meta-Analyses of Perinatal Death and Maternal Exposure to Tobacco Smoke During Pregnancy. Am J Epidemiol. 2016 Jul 15;184(2):87–97.

89. Batstra L, Hadders-Algra M, Neeleman J. Effect of antenatal exposure to maternal smoking on behavioural problems and academic achievement in childhood: prospective evidence from a Dutch birth cohort. Early Hum Dev. 2003 Dec;75(1–2):21–33.

90. Oh SS, Galanter J, Thakur N, Pino-Yanes M, Barcelo NE, White MJ, et al. Diversity in Clinical and Biomedical Research: A Promise Yet to Be Fulfilled. PLoS Med. 2015 Dec 15;12(12):e1001918.

91. Perni UC, Wikström A-K, Cnattingius S, Villamor E. Interpregnancy Change in Smoking Habits and Risk of Preeclampsia: A Population-Based Study. Am J Hypertens. 2012 Mar 1;25(3):372–8.

92. Gudnadóttir TA, Bateman BT, Hernádez-Díaz S, Luque-Fernandez MA, Valdimarsdottir U, Zoega H. Body Mass Index, Smoking and Hypertensive Disorders during Pregnancy: A Population Based Case-Control Study. PloS One. 2016;11(3):e0152187.

93. Fikadu K, G/Meskel F, Getahun F, Chufamo N, Misiker D. Family history of chronic illness, preterm gestational age and smoking exposure before pregnancy increases the probability of

preeclampsia in Omo district in southern Ethiopia: a case-control study. Clin Hypertens. 2020;26:16.

94. Kharkova OA, Grjibovski AM, Krettek A, Nieboer E, Odland JØ. First-trimester smoking cessation in pregnancy did not increase the risk of preeclampsia/eclampsia: A Murmansk County Birth Registry study. PloS One. 2017;12(8):e0179354.

95. Ebbing C, Rasmussen S, Skjaerven R, Irgens LM. Risk factors for recurrence of hypertensive disorders of pregnancy, a population-based cohort study. Acta Obstet Gynecol Scand. 2017 Feb;96(2):243–50.

96. Weinberg CR, Shi M, Basso O, DeRoo LA, Harmon Q, Wilcox AJ, et al. Season of Conception, Smoking, and Preeclampsia in Norway. Environ Health Perspect. 2017 Jun 29;125(6):067022.

97. Lewandowska M, Więckowska B. The Influence of Various Smoking Categories on The Risk of Gestational Hypertension and Pre-Eclampsia. J Clin Med. 2020 Jun 4;9(6):E1743.

98. Ananth CV, Keyes KM, Wapner RJ. Pre-eclampsia rates in the United States, 1980-2010: age-period-cohort analysis. The BMJ. 2013 Nov 7;347:f6564.

99. Lamminpää R, Vehviläinen-Julkunen K, Gissler M, Heinonen S. Smoking among older childbearing women - a marker of risky health behaviour a registry-based study in Finland. BMC Public Health. 2013 Dec 13;13:1179.

100. Jääskeläinen T, Suomalainen-König S, Hämäläinen E, Pulkki K, Romppanen J, Heinonen S, et al. Angiogenic profile and smoking in the Finnish Genetics of Pre-Eclampsia Consortium (FINNPEC) cohort. Ann Med. 2017 Nov;49(7):593–602.

101. Tsiakkas A, Duvdevani N, Wright A, Wright D, Nicolaides KH. Serum placental growth factor in the three trimesters of pregnancy: effects of maternal characteristics and medical history. Ultrasound Obstet Gynecol Off J Int Soc Ultrasound Obstet Gynecol. 2015 May;45(5):591–8.

102. Tsiakkas A, Duvdevani N, Wright A, Wright D, Nicolaides KH. Serum soluble fms-like tyrosine kinase-1 in the three trimesters of pregnancy: effects of maternal characteristics and medical history. Ultrasound Obstet Gynecol Off J Int Soc Ultrasound Obstet Gynecol. 2015 May;45(5):584–90.

103. Zhao H, Wu L, Wang Y, Zhou J, Li R, Zhou J, et al. Nicotine promotes vascular endothelial growth factor secretion by human trophoblast cells under hypoxic conditions and improves the proliferation and tube formation capacity of human umbilical endothelial cells. Reprod Biomed ONLINE. 2017 Apr;34(4):406–13.

104. Wikström A-K, Stephansson O, Cnattingius S. Tobacco use during pregnancy and preeclampsia risk: effects of cigarette smoking and snuff. Hypertens Dallas Tex 1979. 2010 May;55(5):1254–9.

105. Kahn SR, Almeida ND, McNamara H, Koren G, Genest J, Dahhou M, et al. Smoking in preeclamptic women is associated with higher birthweight for gestational age and lower soluble fms-like tyrosine kinase-1 levels: a nested case control study. BMC Pregnancy Childbirth. 2011 Nov 10;11:91.

106. Zhao X, Chen S, Zhao C, Xia F. Maternal Immune System and State of Inflammation Dictate the Fate and Severity of Disease in Preeclampsia. J Immunol Res. 2021;2021:9947884.

107. Venditti CC, Casselman R, Young I, Karumanchi SA, Smith GN. Carbon monoxide prevents hypertension and proteinuria in an adenovirus sFlt-1 preeclampsia-like mouse model. PloS One. 2014;9(9):e106502.

108. Venditti CC, Casselman R, Murphy MSQ, Adamson SL, Sled JG, Smith GN. Chronic carbon monoxide inhalation during pregnancy augments uterine artery blood flow and uteroplacental vascular growth in mice. Am J Physiol Regul Integr Comp Physiol. 2013 Oct 15;305(8):R939-948.

109. Robson A, Harris LK, Innes BA, Lash GE, Aljunaidy MM, Aplin JD, et al. Uterine natural killer cells initiate spiral artery remodeling in human pregnancy. FASEB J Off Publ Fed Am Soc Exp Biol. 2012 Dec;26(12):4876–85.

110. Linzke N, Schumacher A, Woidacki K, Croy B, Zenclussen A. Carbon Monoxide Promotes Proliferation of Uterine Natural Killer Cells and Remodeling of Spiral Arteries in Pregnant Hypertensive Heme Oxygenase-1 Mutant Mice. HYPERTENSION. 2014 Mar;63(3):580–8.

111. Laule CF, Wing CR, Odean EJ, Wilcox JA, Gilbert JS, Regal JF. Effect of nicotine on placental ischemia-induced complement activation and hypertension in the rat. J Immunotoxicol. 2017 Dec;14(1):235–40.

112. Kraus DM, Feng L, Heine RP, Brown HL, Caron KM, Murtha AP, et al. Cigarette smoke-induced placental adrenomedullin expression and trophoblast cell invasion. Reprod Sci Thousand Oaks Calif. 2014 Jan;21(1):63–71.

113. Ghazavi E. The protein expression of the nicotinic acetylcholine receptors in the human placenta, and effects of cigarette smoking and pre-eclampsia. [Internet]. 2014. Available from: https://ses.library.usyd.edu.au/bitstream/handle/2123/10534/GHAZAVI%20Emma%20-%20Final%20thesis.pdf?sequence=1&isAllowed=y

114. He F, Li B, Zhao Z, Zhou Y, Hu G, Zou W, et al. The Pro-Proliferative Effects of Nicotine and Its Underlying Mechanism on Rat Airway Smooth Muscle Cells. PLOS ONE. 2014 Apr 1;9(4):e93508.

115. Nakamura A. Effects of Nicotine Contained in Tobacco Mainstream Smoke on Vascular Smooth Muscle Cells [Internet]. Atherosclerosis - Yesterday, Today and Tomorrow. IntechOpen; 2018 [cited 2021 Dec 5]. Available from: https://www.intechopen.com/chapters/61165

116. Chada A, Lord M, Nguyen H, Davis J, Bouzaher M, Taylor S, et al. Impact of smoking on smooth muscle content of placental vessels in preeclampsia. Am J Obstet Gynecol. 2020 Jan;222(1):S144–S144.

117. Choudhury KM, Das M, Ghosh (Sarkar) S, Bhattacharya D, Ghosh TK. Value of Serum beta-hCG in Pathogenesis of Pre-Eclampsia. J Clin Gynecol Obstet. 2012 Oct 23;1(4–5):71–5.

118. Wright D, Papadopoulos S, Silva M, Wright A, Nicolaides KH. Serum free β-human chorionic gonadotropin in the three trimesters of pregnancy: effects of maternal characteristics and medical history. Ultrasound Obstet Gynecol Off J Int Soc Ultrasound Obstet Gynecol. 2015 Jul;46(1):51–9.

119. Chang JJ, Strauss JF, Deshazo JP, Rigby FB, Chelmow DP, Macones GA. Reassessing the impact of smoking on preeclampsia/eclampsia: are there age and racial differences? PloS One. 2014;9(10):e106446.

120. Nakagawa K, Lim E, Harvey S, Miyamura J, Juarez DT. Racial/Ethnic Disparities in the Association Between Preeclampsia Risk Factors and Preeclampsia Among Women Residing in Hawaii. Matern Child Health J. 2016 Sep;20(9):1814–24.

121. Lisonkova S, Joseph KS. Left truncation bias as a potential explanation for the protective effect of smoking on preeclampsia. Epidemiol Camb Mass. 2015 May;26(3):436–40.

122. Kinlaw AC, Buckley JP, Engel SM, Poole C, Brookhart MA, Keil AP. Left Truncation Bias to Explain the Protective Effect of Smoking on Preeclampsia: Potential, But How Plausible? Epidemiol Camb Mass. 2017 May;28(3):428–34.

123. Wei J, Liu C-X, Gong T-T, Wu Q-J, Wu L. Cigarette smoking during pregnancy and preeclampsia risk: a systematic review and meta-analysis of prospective studies. Oncotarget. 2015 Dec 22;6(41):43667–78.