Altered Maternal and Placental Lipid metabolism and Fetal Fat Development in Obesity: Current knowledge and advances in non-invasive assessment

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Abstract:

Abnormal maternal lipid profiles, a hallmark of increased maternal adiposity, are associated with pregnancy complications such as preeclampsia and gestational diabetes, and offspring long-term metabolic health is impacted as the consequence of altered fetal growth, physiology and often iatrogenic prematurity. The metabolic changes associated with maternal obesity and/or the consumption of a high-fat diet effecting maternal lipid profiles and metabolism have also been documented to specifically affect placental function and may underlie changes in fetal development and life course disease risk.

The placenta plays a critical role in mediating nutritional signals between the fetus and the mother. As obesity rates in women of reproductive age continue to increase, it is becoming evident that inclusion of new technologies that allow for a better understanding of early changes in placental lipid transport and metabolism, non-invasively in maternal circulation, maternal tissues, placenta, fetal circulation and fetal tissues are needed to aid timely clinical diagnosis and treatment for obesity-associated diseases.

This review describes pregnancy lipid homeostasis, with specific reference to changes arising from altered maternal body composition on placental and fetal lipid transport and metabolism. Current technologies for lipid assessments, such as metabolomics and lipidomics may be impacted by labour, mode of delivery, and are only reflective of a single time point. This review further addresses how established and novel technologies for assessing lipids and their metabolism non-invasively and during the course of pregnancy may guide future research into the effect of maternal metabolic health on pregnancy outcome, placenta and fetus.

Keywords: Pregnancy, obesity, lipid metabolism, exosomes, ultrasound, MRI

Highlights

- Maternal lipid status affects placental transport and fetal lipid accumulation
- Obesity is linked to altered placental lipid storage, metabolism and transport
- Maternal exosome analysis may provide new insights in placental function
- US is an established tool to assess fetal adipose tissue relating to fetal growth
- Non-invasive lipid assessments can be done using ^{1}H-MRS (placenta) and MRI (fetus)

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Introduction

Metabolic, inflammatory and endocrine changes in the pregnant mother ensure that the developmental and energy requirements of placenta and fetus are met throughout pregnancy. The adequate supply to the placenta and metabolism of metabolic fuels such as oxygen and carbon dioxide and nutrients such as glucose, amino acids and fatty acids (FA), occur through these changes and are critical to a successful pregnancy outcome [1]. Maternal malnutrition and inadequate weight gain in pregnancy are associated with adverse pregnancy outcomes; however, obesity, over nutrition and excessive gestational weight gain are the more common norms in developed countries, and in recent years, even within developing nations [2]. The growing prevalence of obesity is one of the leading contributors to increasing rates of obstetrical complications, including preeclampsia (PE), gestational diabetes mellitus (GDM), large and small for gestational age fetuses, caesarean section, and iatrogenic preterm birth [3]. For example, maternal hyperlipidemia in the first trimester may be predictive of the later development of PE [4], and a maternal high-fat diet, common in developed countries, is associated with an increased risk of GDM [3]. The altered fetal growth that is the consequence of maternal adiposity and diet-associated obstetrical complications has significant impacts on the health of the offspring later in life [5]. Hence, maternal obesity perpetuates a transgenerational cycle of cardiovascular and metabolic conditions such as offspring obesity, cardiovascular disease and metabolic syndrome (Developmental Origins of Health and Disease (DOHaD) hypothesis) [6]. In this context, the placenta plays a crucial role as it regulates nutrient transfer

from the mother to the fetus. Metabolic, endocrine and inflammatory changes associated with maternal obesity/high-fat diet affect placental function and, in turn, induce changes in fetal development.

Current techniques to assess placental and fetal fat require post-delivery examinations of blood and tissue after the disease processes have already advanced and the postnatal metabolic health outcome is potentially programmed [7]. Furthermore, lipid levels may be affected during labour, making it difficult to obtain reliable data on placental and fetal fat metabolism [8, 9]. Investigating changes in placental and fetal lipid metabolism and function in early pregnancy could allow for early detection of abnormal fetal growth and prevent future metabolic and developmental complications in offspring. New technologies are emerging that now allow for the investigation of changes in lipid metabolism, non-invasively in all compartments in maternal circulation, maternal tissues, placenta, fetal circulation and fetal tissues [10-16].

This review describes normal pregnancy lipid homeostasis, known effects of obesity and intake of high energy diets on placental and fetal lipid transport and metabolism, and how techniques for assessing placental and fetal lipid profiles and metabolism may guide future clinical practise and research into the effect of maternal obesity/overnutrition on placenta and fetus.

Impact of abnormal maternal lipid profiles on placental lipid transport and fetal development

Maternal lipid metabolism during an uncomplicated pregnancy is divided into an anabolic and a catabolic phase [17]. The anabolic phase occurs during the first two trimesters and is associated with increased storage of maternal FA, triglycerides (TG), phospholipids (PL),

cholesterol and lipoproteins (LP) to ensure a continuous supply of nutrients to the growing fetus later in pregnancy [1, 18]. During this anabolic phase, increased levels of maternal estrogen, progesterone, and insulin promote lipogenesis and inhibit lipolysis [1] leading to significant increases in plasma lipid concentrations throughout gestation [19]. Circulating TG are hydrolyzed by the enhanced activity of adipose tissue lipoprotein lipase (LPL) resulting in increased uptake of non-esterified fatty acids (NEFA) in maternal tissues [20]. The catabolic phase of maternal lipid metabolism begins in the third trimester and involves increased lipolysis and decreased LPL activity in adipose tissue initiating fat breakdown and the release of NEFA, PL and cholesterol into the maternal circulation for transport to the growing fetus [17, 20]. As pregnancy advances, fasting glucose levels decline while hepatic glucose production and fasting insulin levels rise, resulting in decreased insulin sensitivity [21]. Placental growth hormone (GH-V) is involved in inducing this maternal insulin resistance [22] and adipokines, such as leptin and adiponectin, also play a role in the progression of insulin resistance in normal pregnancies, to ensure availability of carbohydrates to meet the metabolic needs required for pregnancy [23, 24]. Additionally, increasing placental lactogen induces maternal leptin resistance and aids in β-cell expansion to promote insulin production to prevent the development of hyperglycemia in healthy pregnancies [22]. A systemic inflammatory response is also required for normal pregnancy progression, beginning with implantation and placentation through to parturition [25]. Pro-inflammatory cytokines, such as IL-6 and TNF-α, have shown to play a role in regulating placental transport through the activation of system A, which is responsible for transport of essential and non-essential amino acids across the placenta [26]. Together, these physiologic changes ensure that the placenta and its fetus receive a continuous and targeted supply of the

appropriate FA species; for membrane development in the first trimester and for fat accretion and metabolism in the third trimester.

The placenta is the primary facilitator for nutrient transfer from mother to fetus and is essential for adequate fetal growth. It regulates the availability of FA, adapting to constant changes in demands for the developing fetus as well as for itself. It does so through either facilitated transport, with the help of membrane transport proteins, or simple diffusion, regulated by the concentration gradient between the maternal and fetal circulations [27-30]. For instance, the placenta regulates the transport of long chain polyunsaturated FA (LCPUFA), docosahexaenoic acid (DHA) and arachidonic acid (ARA) in particular as these are critical components for fetal development and cannot be produced by the human body [31]. This process, termed "biomagnification", involves the selective increase of LCPUFA in the fetal circulation as gestation progresses [31]. Similar to most tissues, but unlike adipose tissue and liver, the placenta has limited or no capacity for FA synthesis and requires a steady maternal supply to meet its needs. Figure 1A outlines the supply and transport of lipids from the maternal circulation into the placenta, with key aspects of placental metabolism and storage as well as the movement of FA across the placenta to the fetal circulation [26-30, 32-37].

Women of reproductive age often struggle to comply with nutrition and lifestyle recommendations for planning a pregnancy [38] and often exceed the modest increase in dietary energy intake of 375, 1200 and 1950 kJ per day required for the 1st, 2nd and 3rd trimester of pregnancy, respectively. In addition, pregnant women are advised to aim for a dietary intake of the essential n-3 LCPUFA that supplies a DHA intake of at least 200 mg/d or the equivalent of two portions of fatty fish per week, but no evidence exists to support a change in the total dietary

fat intake as a percentage of total caloric intake [39]. As a result, many have imbalanced levels of the essential LC-PUFAs including DHA and ARA [38].

Women with obesity generally consume larger amounts of saturated FA [40] and have a smaller proportion of metabolically active organs such as muscle, relative to their body size [41]. These obesity-related changes are associated with abnormal maternal lipid profiles during pregnancy, with higher circulating content of TG and cholesterol compared to pregnant women with a normal body mass index (BMI) [42]. Additionally, women who are overweight or obese have elevated concentrations of NEFA in the third trimester [43].

Obesity is also associated with alterations in inflammation and endocrine function. These processes are highly regulated throughout gestation for adequate placental and fetal development [25, 44]. Increased levels of placental inflammatory markers such as IL-6, IL-8, and TNF-α have been noted in pregnancies affected by obesity [45], which may play a role in programming fetal adipose tissue, liver and skeletal muscle for insulin resistance later in life [45]. A lower adiponectin/leptin ratio, a marker of insulin resistance, has also been observed in obese pregnancies [23], and has been associated with a higher fetal weight [46]. These altered maternal levels of adipokines have been associated with increased insulin resistance and percent body fat in the fetus as well as increased adiposity at 3 years of age [47, 48]. In maternal obesity, the placenta displays increased lipid esterification and storage, in conjunction with decreased mitochondrial fatty acid oxidation (FAO) and increased peroxisomal FAO [49].

Metabolomic studies in placentae collected after elective Caesarean section have established that obesity is associated with higher placental lipid accumulation and metabolism [50]. In addition, pre-pregnancy obesity is associated with decreased placental saturated FA content [51] as well as a disruption of LCPUFA biomagnification, leading to decreased

availability of AA and DHA to the developing fetus [50]. Furthermore, increased placental mTOR signaling has been reported in obese women delivering large babies and in experimental models of maternal obesity associated with fetal overgrowth [52], highlighting the important role of mTOR signaling in regulating growth and development through increased nutrient transporter expression and function [53, 54]. These results provide insight into the mechanisms of how elevated maternal FA concentrations may impact placental function and adversely regulate fetal exposure to FA (Figure 1B).

Maternal obesity has been associated with elevated levels of TG in the fetal circulation [55]. Both obesity and obesity-related GDM display increased plasma leptin and insulin levels in cord blood [56, 57] and particularly high fetal insulin drives metabolic pro-adipogenic changes, including hepatic FA and TG synthesis in white adipose tissue [35]. These changes predispose the fetus to increased fat deposition, higher birth weight and postnatal adiposity [58].

Human cadaver studies of normally grown fetuses have identified fetal adipose tissue to develop as early as 14 weeks gestational age, first in head and neck, somewhat later in the thorax and abdomen, and finally in extremities [59], with no significant differences related to fetal sex in any of the fat deposits. Mature adipocytes, composed of approximately 90% lipid contained in a single large vacuole [60], develop from preadipocytes with multiple vacuoles [61].

In mammals, two main types of adipose tissue exist; white adipose tissue (WAT) and brown adipose tissue (BAT). WAT is mainly composed of a single lipid-filled vacuole and functions in energy storage, whereas BAT contains multiple lipid vacuoles, uncoupling protein-1 (UCP1) and an abundance of mitochondria and functions in thermogenesis [62]. The ontogeny of BAT is species-dependent; BAT recruitment for large mammals (i.e. humans) is greatest just prior to birth whereas in smaller mammals (i.e. rodents), the development of BAT is most

abundant after birth [62]. Most mammals accumulate very little adipose tissue until after birth, whereas human infants require adipose tissue to endure the stresses of labour and to promote thermoregulation after birth [62, 63]. During fetal development, the distinction between the two types of adipose tissue is difficult to make, as both developing WAT and BAT show a multivacuolated phenotype with a very similar appearance on histology without using immunohistochemical markers for thermogenic tissues (UCP1) or specific for genes overexpressed specifically in BAT (PRDM16, PGC1α, and RXRγ) [64]. For this reason, the developmental timing of WAT and BAT development remains relatively understudied. A distinction between WAT, considered disadvantageous for the developing fetus as it predisposes to obesity and metabolic syndrome (MetS) [65], and the thermoregulatory BAT essential for survival in a cold environment [66] immediately after birth, may be essential in providing information about fetal metabolic health and future disease risk.

Maternal BMI has been shown to be directly correlated with neonatal weight [67], a higher rate of fat deposits [68] as well as an increased risk of obesity and type 2 diabetes later in life [5]. Similarly, increased total and percent fat mass and decreased fat-free mass have been found in infants of overweight and obese women [69, 70] and subcutaneous increase of WAT deposits under the skin and/or around the internal organs are associated with increased health risks in adult life [65]. The effect of increased availability of FA to fetal circulation extends to other metabolic and endocrine organs, such as maternal liver and muscle. In response to increased availability of nutrients to fetal circulation, the partitioning of blood flow to the fetal liver increases, resulting in increased levels of growth factors contributing to fetal size and fat mass [71, 72], and likely contributing to the development of non-alcoholic fatty liver disease (NAFLD) and/or MetS in the offspring [73, 74].

Established and novel techniques for assessing placental and fetal lipids *in utero*

Although biological techniques such as lipidomics and metabolomics have provided most of the current knowledge of lipid metabolism, they do not allow for analysis of the physiologic state of pregnancy. Assessments of these tissues after birth only provides a snapshot of a single time point and does not consider the changes that occur due to preoperative fasting and/or labour. For example, when compared to patients receiving a preoperative carbohydrate drink, those who fasted prior to surgery had increased circulating NEFA levels [8]. Similarly, labour itself is associated with increases in maternal oxidative stress and inflammation, as well as elevated levels of free FA in the maternal circulation [8, 75, 76]. To address these concerns, several studies have attempted to assess placental and fetal fat *non-invasively*, throughout gestation with various techniques, including exosome analysis, proton magnetic resonance spectroscopy (^{1}H-MRS), ultrasound, and magnetic resonance imaging (MRI) (Figure 2). Advantages and disadvantages of each of these techniques are outlined in Table 1.

It is evident that altered placental lipid homeostasis has developmental and metabolic effects on the fetus *in utero*. Therefore, two novel non-invasive technologies, exosome analysis and ^{1}H-MRS, have been developed to assess placental lipid metabolism throughout gestation. Exosomes are small (40 - 120 nm), extracellular vesicles that are formed within cells and, therefore, their contents represent the state of the cell in which they were produced [77]. They contain a variety of signalling molecules such as protein, mRNA, microRNA, and noncoding RNA [78], and are released into the maternal circulation by placental syncytiotrophoblast cells in response to environmental changes such as oxygen tension and altered glucose concentration

[79]. This release occurs as early as six weeks gestation [78] making it possible to analyze placental composition over the entire course of the pregnancy.

A plethora of proteins have been detected within exosomes as early as the first trimester, including membrane transporters such as apolipoproteins, LDL-receptor related protein 1, and pro-inflammatory cytokines such as tumour necrosis factor and IL-26 [79]. Exosomes derived from syncytiotrophoblast cells have been used to determine changes in the placental lipid profile in pregnancies complicated by PE and recurrent miscarriages. Sphingomyelin was found to be the most abundant lipid in placental exosomes, followed by cholesterol, phosphatidylcholine, phosphatidylserine and phosphatidylinositol [80], and both PE and recurrent miscarriages were associated with increased levels of phosphatidylserine and decreased levels of phosphatidylinositol, phosphatidic acid, and ganglioside mannoside 3 [80].

Exosomes have not been extensively studied in pregnancies affected by maternal obesity or abnormal fetal growth. Exosome numbers in maternal circulation are increased in obese pregnancies, potentially a reflection of elevated lipid levels, inflammation, hyperglycemia and oxidative stress [12]. It is suggested that exosomes are involved in maternal-fetal immuno-tolerance, maternal systemic inflammation and nutrient transport [81]. Although further research is still required to standardize protocols for the acquisition and assessment of extracellular vesicles such as exosomes [78], measurement of lipid content as well as genes and proteins involved in lipid transport and metabolism may provide new insights into placental function.

The placenta is composed of very little lipid (~1%), with water and protein accounting for approximately 88% and 11% of the total weight, respectively [82]. Lipidomics techniques have been used to measure relative lipid contents after delivery of the placenta, and have shown that placental lipids consisted of approximately 60% PL, 34% cholesterol, 4% cholesterol esters

and 2% TG [83]. This small amount of lipid in the placenta makes it difficult to measure with imaging techniques *in utero*, although some studies have attempted this using ^{1}H-MRS. ^{1}H-MRS non-invasively measures relative concentrations of metabolites in a volume of interest [10]. Limitations of ^{1}H-MRS include low spatial resolution (typical voxel size greater than 1 cm^3), limited spatial coverage (typically single voxel acquisition), and lengthy acquisition times. Additionally, access to magnetic resonance facilities is limited and ^{1}H-MRS is not routinely used or widely available clinically. Studies using ^{1}H-MRS found no differences in the concentrations of lipids in placentas from small for gestational age (SGA) pregnancies [10, 84], but demonstrated a significantly reduced choline/lipid concentration ratio in placentas associated with severe intrauterine growth restriction (IUGR) [10]. It is also possible to characterize lipid structure using ^{1}H-MRS, and this technique can be used to distinguish types of lipids [85]. However, this technique has not yet been used to assess changes in placental lipid metabolism, transport or storage that may be associated with maternal obesity.

Current understanding of fetal lipids throughout gestation is limited. Recent publications have demonstrated that imaging techniques such as ultrasound and MRI can be used to assess the development of fetal lipids. Due to the low lipid content of most other tissues, these imaging measurements have been restricted to adipose tissue. Estimating fetal adipose tissue volume *in utero* is most commonly done using ultrasound because it is readily accessible and has a fast acquisition time. Limitations of ultrasound include operator-dependent data acquisition, limited field of view, limited availability of 3D acquisition, and only indirect measurement of lipids using fetal adipose tissue thickness. Multiple studies have used ultrasound measurements of subcutaneous tissue thickness to identify abnormal fetal growth. These techniques have, for instance, identified correlations between fetal thigh subcutaneous tissue thickness and SGA and

large for gestational age (LGA) infants [13], between abdominal wall subcutaneous tissue thickness and prediction of fetal growth restriction [14], and persistence of increased scapular and abdominal subcutaneous tissue thickness in fetuses of diabetic mothers into the neonatal period [15]. Ultrasound measurement of fetal subcutaneous tissue area, by subtracting lean area including muscle and bone from total fetal area, has been used in the prediction of birth weight [86] and neonatal adiposity [87]. Volumetric measurements of fetal tissues obtained using three-dimensional ultrasound [88] is a more complex technique used to estimate birth weight and neonatal fat mass that has not been shown to be superior to the simpler measurement of subcutaneous tissue area [88].

MRI has been demonstrated to be safe for both the mother and fetus [89] and provides volumetric measurements to estimate fetal weight with an accuracy superior to ultrasound estimations [16]. Unlike ultrasound techniques, MRI can image the entire fetus and is able to measure lipid content, rather than adipose tissue volume only. Limitations of MRI include technical issues such as lengthy acquisition times resulting in sensitivity to motion, and logistic issues such as limited access to magnetic resonance facilities and limited clinical availability. A specific limitation related to lipid quantification with MRI is that, while mobile lipids are visible, those lipids contained in structures such as the phospholipid bilayer and myelin are extremely challenging to detect.

There are a variety of MRI techniques that can be used for imaging fetal adipose tissue. T_1-weighted MRI distinguishes different tissues based on their T_1 (an intrinsic magnetic resonance tissue property). Adipose tissue has a shorter T_1 relative to most other tissues and therefore appears brighter than most other tissues in T_1-weighted images [90]. Water-suppressed MRI uses frequency-selective radiofrequency pulses to suppress signal from water and therefore

generates images of only lipid-containing structures [91]. Chemical-shift encoded MRI (CSE-MRI) uses the frequency difference between water and lipid to simultaneously produce water-only and fat (lipid) only images [90]. While all of these techniques produce images of lipid distribution, they are subject to biases that prevent quantification of lipid content [92]. Quantitative CSE-MRI accounts for biases in the MRI signal, including but not limited to T_1, T_2^*, and lipid spectral complexity, to produce images of quantitative lipid content using a parameter referred to as proton density fat fraction (PDFF) [92].

Using T_1-weighted images acquired during a maternal breath hold [91, 93-95], the fetal subcutaneous adipose tissue layer can be easily measured from 29 weeks gestation [95] and allows for differentiation between small fetuses who later developed significant neonatal morbidities from those with no postnatal complications [93]. While decreased fetal adipose tissue volume is associated with IUGR [94], fetuses from diabetic mothers have greater adipose tissue accumulation in the third trimester [91]. One study used MRI to assess the development of fetal adipose tissue from 23-37 weeks gestation by comparing the signal intensity of adipose tissue to that of muscle tissue [96]. This study found that the signal of subcutaneous fetal adipose tissue became relatively more intense with increasing gestational age, suggesting that fetal adipose lipid content is increasing as pregnancy advances.

Quantitative CSE-MRI is a promising technique for non-invasive measurement of fetal adipose tissue. As adipocytes develop, they accumulate larger lipid vesicles; a change that can be measured using CSE-MRI. Neonatal CSE-MRI studies found that the PDFF of infant WAT is lower (60-90%) than that of adults (90%), suggesting that neonatal subcutaneous fat is still in the process of accumulating free lipids [97]. Preliminary studies in the third trimester of human pregnancy show that the PDFF in fetal subcutaneous adipose tissue rapidly increases with

gestational age [98]. This highlights the utility of this technique for assessment of fetal lipid storage development in both normal and abnormal pregnancies and in relation to future development of adiposity. Finally, in neonates, MRI has demonstrated the ability to noninvasively distinguish between BAT and WAT, with a significantly lower fat fraction in BAT (38.2%) than in WAT (77.9%) [97]. Whether this distinction can be made *in utero* depends on whether there are differences in lipid content between BAT and WAT during fetal adipose tissue development.

Conclusions

A normal pregnancy is characterized by both an elevated maternal lipid profile and increased insulin resistance. As the fetus develops, it obtains all of its nutrients from the mother through the placenta. The placenta selectively transports FA according to the demands of the fetus as well as its own metabolic needs. Transport across the placenta is highly regulated through several different mechanisms. Alterations to these mechanisms may hinder or accelerate fetal growth (Figure 1).

The rise in obesity parallels the increased rate of obstetric complications such as PE, GDM, LGA/macrosomia and SGA/IUGR. In obesity and over-nutrition, higher concentrations of lipids in the maternal circulation cross the placenta and enter the fetal circulation. Abnormal lipid transport across the placenta results in improper fetal development and metabolic abnormalities in the offspring. Altered placental transport may be due to changes in the expression of fatty acid transport proteins (FATPs), placental fat storage, or nutrient sensing through the mTOR pathway. Hormones such as leptin and insulin also appear to be important in fetal growth and

development. Elevated levels of these hormones in the fetal circulation, associated with maternal obesity and overnutrition, may increase fat storage and affect the metabolism of the offspring.

The ability to measure fat *in utero* as it is transported from mother to fetus would aid in understanding the connection between maternal health and pregnancy outcomes. Current methods to evaluate changes to lipids in pregnancy are generally limited to the postpartum period. Novel methods are now emerging that can be applied throughout gestation to gain a better understanding of maternal, placental and fetal lipids (Figure 2). These methods may also be useful for earlier detection of improper fetal growth, allowing for potential interventions such as dietary supplementation or modifications. Novel techniques have shown promise in their ability to measure lipids as they move from the maternal circulation, across the placenta and into the fetal circulation. For instance, syncytiotrophoblast-derived exosomes may be reflective of the lipids in the placenta, and fetal fat development may be assessed non-invasively using CSE-MRI. Further research using existing non-invasive techniques and incorporating newly-developed technologies is crucial in improving our understanding of both normal and abnormal physiology. This will help to overcome the retrospective nature of postnatal studies such as cord blood and placental analyses that may be impacted by labour and mode of delivery, and are only reflective of a single time point.

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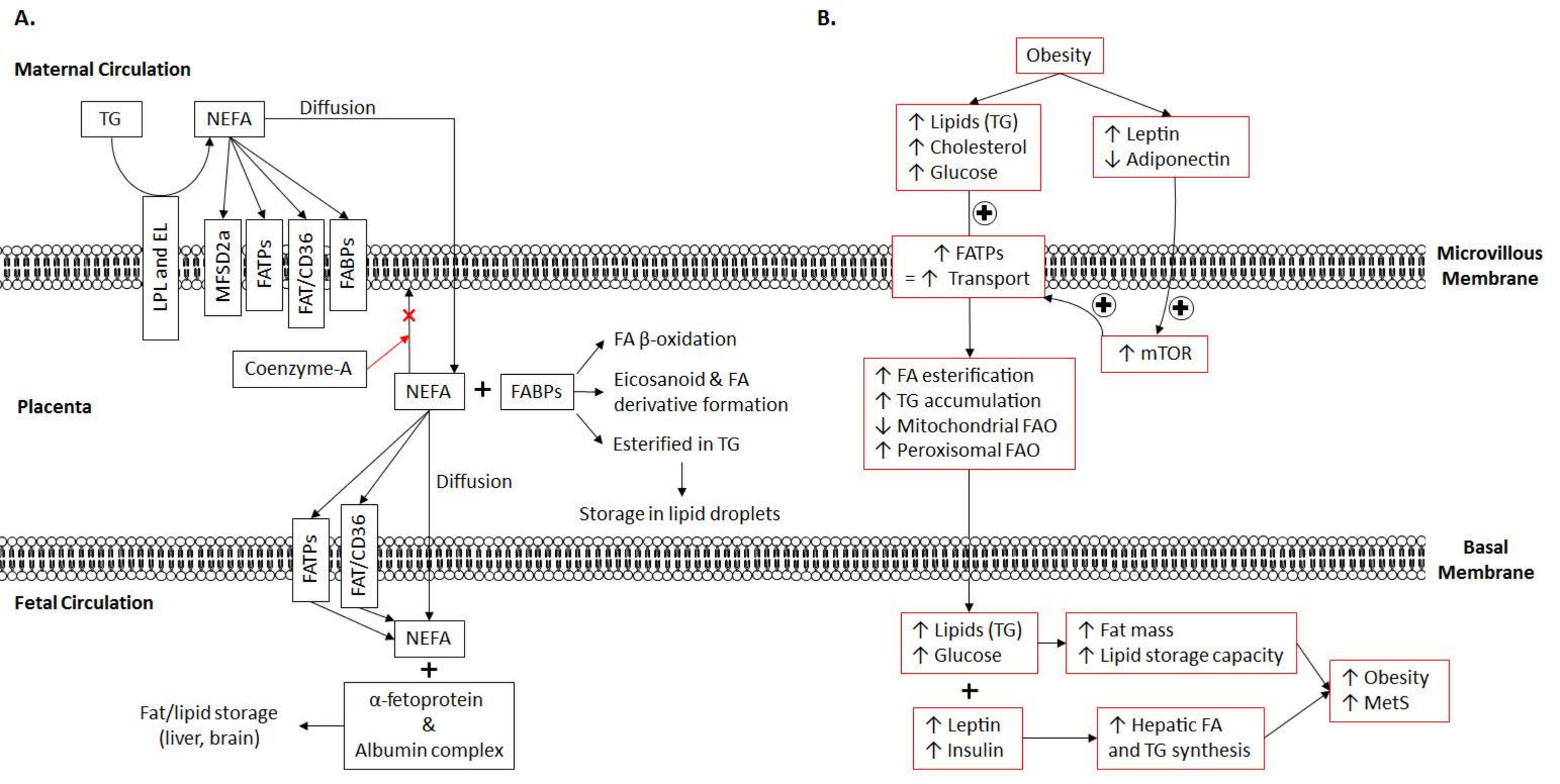
A.
Maternal Circulation
TG
NEFA
Diffusion
LPL and EL
MFSD2a
FATPs
FAT/CD36
FABPs
Coenzyme-A
NEFA
+
FABPs
FA β-oxidation
Eicosanoid & FA derivative formation
Esterified in TG
Storage in lipid droplets
Placenta
Diffusion
FATPs
FAT/CD36
Fetal Circulation
NEFA
+
α-fetoprotein & Albumin complex
Fat/lipid storage (liver, brain)
B.
Obesity
↑ Lipids (TG)
↑ Cholesterol
↑ Glucose
↑ Leptin
↓ Adiponectin
↑ FATPs
= ↑ Transport
↑ mTOR
Microvillous Membrane
↑ FA esterification
↑ TG accumulation
↓ Mitochondrial FAO
↑ Peroxisomal FAO
Basal Membrane
↑ Lipids (TG)
↑ Glucose
↑ Fat mass
↑ Lipid storage capacity
+
↑ Leptin
↑ Insulin
↑ Hepatic FA and TG synthesis
↑ Obesity
↑ MetS

Figure 1: Impact of obesity on placental lipid transport and metabolism and fetal development

A. *Unaltered placental lipid transport and metabolism*

FA must first cross the microvillous membrane facing the maternal circulation and then the basal membrane on the fetal side to reach the fetal circulation. LPL and endothelial lipase (EL) on the maternal surface of the syncytiotrophoblasts hydrolyze maternal TG to release NEFA into the cytosol. NEFA are able to cross the placenta either by simple diffusion driven by the concentration gradient from mother to fetus, as well as facilitated diffusion controlled by membrane transport proteins such as FATPs, fatty acid translocase (FAT/CD36), fatty acid binding proteins (FABPs) and the MFSD2a transporter. Once in the cytosol, FABPs act to facilitate cytosolic trafficking to sites for processing. FA are ligated by coenzyme-A forming acyl-CoA where they can be oxidized through β-oxidation, converted into eicosanoids, or esterified in TG and PL and stored in lipid droplets. The FA that are not stored or utilized by the placenta are then transported across the BM into fetal circulation through both facilitated and simple diffusion. Once in the fetal circulation, FA bind to either α-fetoprotein or albumin to reach the fetal liver.

B. *Altered placental lipid transport and metabolism*

Placental transport and metabolism of lipids is affected by obesity and its related complications, with altered expression of FATPs, increased activity of the mTOR signalling pathway, increased lipid esterification and storage, decreased mitochondrial FAO and increased peroxisomal FAO in the placenta. The increased concentrations of lipids (i.e. TG) in fetal circulation could result in improper fetal development and elevated levels of leptin and insulin may impact fetal fat storage and affect the metabolism of the offspring later in life (obesity, MetS).

Mother

↑ obesity

↓

Endocrine Function:
↓ leptin/adiponectin
↑ insulin resistance

Lipid Profile:
↑ lipids (TG)
↑ cholesterol
↑ glucose

Lipidomics
Maternal lipid concentrations

Placenta

Lipid Transport:
↑ FATP
↑ mTOR

Metabolism/Storage:
↑ FA esterification
↑ TG accumulation

^{1}H-MRS
Relative lipid concentrations

Exosomes
Placental lipid content

Fetus

Endocrine Function:
↑ leptin
↑ insulin

Metabolism/Storage:
↑ hepatic FA and TG synthesis
↑ fat mass/storage

Lipid Profile:
↑ Lipids (TG)
↑ glucose

Ultrasound
Tissue thickness/area/volume

MRI
Fetal lipid content

Figure 2: Non-invasive techniques for assessing *in utero* lipid transport and metabolism

Figure 2 highlights where each non-invasive technique may be used to assess lipid transport and metabolism throughout gestation (i.e. mother, placenta or fetus) as well as what each technique can measure. These measurements can be realized throughout gestation in both physiological and complicated pregnancies, unaffected by labour or birth.

Table 1: Comparing techniques measuring placental and fetal lipids throughout gestation

Technique	Advantages	Disadvantages
^{1}H-MRS (in the placenta)	• Can measure very small amounts of lipids	• Very slow acquisition (sensitive to motion) • Low spatial resolution • Limited accessibility
Exosomes (released from placenta to maternal circulation)	• Easy to collect (maternal blood sample) • Reflect placental composition	• Lack of standardized protocols available
Ultrasound (of the fetus)	• Readily accessible • Very fast acquisition	• Limited to measuring thickness/area/volume • Technician dependent • Not directly sensitive to lipid (cannot measure lipid content)
MRI (of the fetus)	• Can image entire fetal volume/uterus • Directly sensitive to lipid (can measure lipid content through water-fat MRI)	• Slow acquisition (sensitive to motion) • Limited accessibility

Table 1 lists advantages and disadvantages of using each technique to non-invasively assess placental and fetal lipids *in utero*.