Altered foetal-placental vascular endothelial signalling to insulin in diabesity

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\textbf{A B S T R A C T}

Obesity and type 2 diabetes mellitus (T2DM) are diseases associated with hypertension and metabolic alterations. A significant group of patients present both obesity and T2DM, a condition defined as diabesity. One of the metabolic features in these conditions is the clinical presentation of insulin resistance. Several tissues, including the liver, skeletal muscle, and vasculature, and patients with T2DM, gestational diabetes, and obesity show insulin resistance. The vascular effect of insulin, including vasodilation, is mainly mediated by the generation of nitric oxide. Several mechanisms are proposed to elucidate the origin of insulin resistance; nevertheless, a common finding is the endothelial dysfunction in these diseases. Endothelial cells from subjects with obesity show reduced nitric oxide synthesis, an effect that is unaltered by insulin. Individuals with T2DM show a misbalance between the synthesis, release, and biological actions of vasodilators and vasoconstrictors such as nitric oxide and endothelin-1. However, whether these mechanisms are involved in the vascular alterations seen in patients with diabesity is unclear. In this review, we discussed the modifications on insulin signalling, insulin resistance in obesity and T2DM, and the reported changes in signalling pathways in diabesity.

1. Introduction

Obesity results from an imbalance between the energy intake and energy expenditure ending in higher adipose tissue volume due to an over-storage of lipids in the adipocytes (Shoelson et al., 2007; World Health Organization (WHO), 2018). Obesity relates to metabolic alterations including hypertension, dyslipidaemia, insulin resistance, and type 2 diabetes mellitus (T2DM) (Fernández-Sánchez et al., 2011; Ouchi et al., 2011). This pathology also associates with macrovascular and microvascular endothelial dysfunction and a higher incidence of cardiovascular disorders (Ianotto et al., 2014; Lobato et al., 2012; Pardo et al., 2015, 2017, 2018; Tesauro and Cardillo, 2011). Obesity and insulin resistance are also seen in individuals with T2DM, a metabolic disease whose worldwide prevalence is increasing (WHO, 2018). Diabetes mellitus could be of type 1 (T1DM) or T2DM. T1DM results from blunted insulin synthesis due to pancreas β-cells autoimmune destruction (ADA, 2018; WHO, 2018). T2DM is regarded as a progressive insulin secretory defect on the background of insulin resistance with increased hyperglycaemia, reduced insulin receptor sensitivity, and defective insulin-triggered signalling in insulin target tissues (ADA, 2018; WHO, 2018).

An increase in the worldwide prevalence of obesity and T2DM is seen in the last decade (ADA, 2018). The combination of these two pathological conditions, with or without associated risk factors, is
defined as diabesity (Kalra, 2013; Ziv and Sharif, 1995). One of the effects of diabesity in the cardiovascular system includes worsening of the vascular response to insulin and other vasodilators. It is now well documented that obesity, T2DM, and diabesity are pathological conditions associated with a subnormal biological response to insulin, i.e. insulin resistance, which is seen as a broad spectrum of alterations in the metabolic state of the cells including a subnormal glucose response (Di Meo et al., 2017; Lebovitz, 2001; Moller and Flier, 1991). Worryingly, there is an increase in the prevalence of obesity in women in their childbearing age (Araya et al., 2004; National Health Survey (ENS, 2017; Centers for Disease Control and Prevention (CDC, 2017)). Also, an association between pre-gestational maternal obesity and gestational diabetes mellitus (GDM), a disease that appears in the second or third trimester of pregnancy, is reported (Bider-Canfield et al., 2017; Chu et al., 2007). Thus, a more significant number of women developing ‘gestational diabesity’ (i.e. maternal obesity + GDM) in the coming years is expected (Pardo and Sobrevia, 2018; Cabalín et al., 2019). This review summarises the general characteristics of insulin resistance in obesity and diabesity. The possibility that insulin resistance may be a different metabolic condition in gestational diabesity is also discussed.

2. Insulin signalling and insulin resistance

2.1. Normal insulin signalling

Insulin is a hormone whose primary function is to regulate the metabolism of D-glucose and other metabolic substrates, such as amino acids and lipids, in target tissues (Silva et al., 2017; Tokarz et al., 2018; Villalobos-Labra et al., 2017). The biological effects of insulin are mediated by the activation of insulin receptors of which at least two splice variants are reported, i.e. insulin receptor A (IR-A) and B (IR-B) (Westermeier et al., 2016). Both IR-A and IR-B are expressed in the vascular endothelium and smooth muscle, including the human placenta and umbilical cord vessels (hereafter referred as ‘foeto-placental vasculature’) (Westermeier et al., 2011; Subiabre et al., 2017).

Activation of IR-A is associated with a preferential mitogenic compared with a metabolic phenotype induced by activation of IR-B in the vascular endothelium (Solomón et al., 2012; Westermeier et al., 2016) (Fig. 1). The affinity of IR-A for insulin is higher compared with IR-B (Mostaf et al., 1990; Westermeier et al., 2016) and activation of these variants results in activation by β-subunit autophosphorylation (Boucher et al., 2014; Westermeier et al., 2015). The autophosphorylation of the insulin receptors leads to recruiting and causing activator phosphorylation of insulin receptor substrates (IRSs) and the Src homology 2 domain-containing transforming protein 1 (SHc) (Boucher et al., 2014). IR-A activation results in preferential but not exclusive activation of IRS-1 and IRS-2 which are members of a family group formed by at least six identified members (IRS-1 to IRS-6). It is well established that activation of IR-A/IRS-1 and IR-A/IRS-2 increases the activity of phosphatidylinositol 3 kinase (PI3K) with minor activation of the 44 and 42kDa mitogen-activated protein kinases (p44/42MAPK) (Ravichandran et al., 2001). On the other hand, insulin acting on IR-B causes preferential activation of three SHc proteins, i.e. SHcA, SHcB, and SHcC, of which the SHcA shows alternative splicing isoforms in mammals (SHcA 46, SHcA 52, and SHcA 66) (Ravichandran et al., 2001). The IR-B/SHcA preferential activation in response to insulin results in higher activation of p44/42MAPK compared with protein kinase B/Akt (Akt) via the growth factor receptor-bound protein 2 (Grb2) (Ong et al., 2001). Both the IR-A/IRSs and IR-B/SHcA signalling target the nitric oxide (NO) synthase (NOS) leading to higher generation of NO via the endothelial (eNOS), inducible, and neuronal NOS (Subiabre et al., 2017; Kellogg et al., 2017) in most, if not all, cell types including the vascular endothelium. In healthy subjects, insulin increases the NO generation by preferential activation of IR-B signalling in the endothelium leading to relaxation of the underlying smooth muscle and vasodilatation (Fig. 2).

The mechanisms behind the insulin effects in the vasculature include activation of L-arginine transport, the substrate of NOS (Fleming, 2010), as a result of a higher expression and maximal transport capacity of the cationic amino acid transporters 1 (hCAT-1) and hCAT-2B (apparent *Km* 50–250 μmol/L) (González et al., 2015; Subiabre et al., 2017). Activation of the L-arginine transport via hCAT-1/hCAT-2B (González et al., 2015; Mann et al., 2003) and system y’+L activity (high affinity transporters, apparent *Km* 1–10 μmol/L) (Ramírez et al., 2018) is proposed to increase the NO generation in several cell types including the vascular endothelium. Insulin-increased hCAT-1/hCAT-2B transport activity resulted from higher expression of the SLC7A1 and SLC7A2 (a high affinity spliced variant of SLC7A2) due to activation of p44/42MAPK in human umbilical vein endothelial cells (HUVECs) (González et al., 2015). Insulin effect required the activation of the transcription factor specific protein 1 to increase the expression of SLC7A1 in this cell type.

2.2. Defective insulin signalling

When the signalling mechanisms at any level are defective after activation of insulin receptors, a condition of insulin resistance is seen in most cells (Lebovitz, 2001; Moller and Flier, 1991; Silva et al., 2017; Villalobos-Labra et al., 2017). Insulin resistance is a condition that could initiate in individuals showing functional pancreas β cells thus releasing insulin in response to altered D-glucose, lipids and protein metabolism, i.e. individuals with diabetes mellitus referred to as non-insulin dependent subjects (Di Meo et al., 2017). The altered response to insulin is first seeing as a reduced response to insulin in the primary target tissues to this hormone, i.e. the skeletal muscle, the adipose tissue, and liver (Villalobos-Labra et al., 2018b). Obesity (Yazici and Sezer, 2017; Villalobos-Labra et al., 2018b; Cabalín et al., 2019), sedentarism (Di Meo et al., 2017; Hamilton et al., 2014), and genetic background (Feng et al., 2019) are significant risk factors for developing insulin resistance. However, insulin resistance occurs in people that show hyperinsulinemia, excessive fat stored in the liver and pancreas, and high levels of inflammation. Most studies regard insulin resistance as due to an abnormal D-glucose uptake and metabolism in large organs such as the skeletal muscle. However, the precise mechanisms for this abnormal metabolic condition are still unclear.

Insulin resistance is shown to associate with activation of MAPK signalling pathways in vascular cells and other cell types (Westermeier et al., 2016). The latter is a phenomenon that may result from a higher expression of IR-A in the vascular endothelium (Villalobos-Labra et al., 2017; Westermeier et al., 2016). In a pathophysiological environment, insulin increases the endothelin 1 (ET-1) secretion in a MAPK activation-dependent manner resulting in vasoconstriction (Cardillo et al., 1999) (Fig. 2). Since the endothelin receptor A is the predominant receptor isoform for ET-1 expressed in vascular smooth muscle cells (Thorin and Webb, 2010), ET-1 may act as a vasoconstrictor via activation of this type of receptors in insulin resistance as described in bovine vascular smooth muscle cells (Arai et al., 1990). ET-1 also leads to NO-dependent vasodilatation acting on endothelin receptor B2 expressed in the endothelium (Sakurai et al., 1990). Thus, insulin-increased ET-1 secretion could also increase the NO synthesis in endothelial cells. However, ET-1 biological action will depend on the distribution pattern of endothelin receptors in the vascular endothelium and smooth muscle. NO also inhibits ET-1 biological effect by reducing this molecule-vasoconstriction favouring vasodilatation (Rapport et al., 2014).

Human pregnancy is a physiological condition where the mother and foetus show with increased plasma levels of insulin in response to the increased substrate demand by the foetus (Sobrevia et al., 2015; Subiabre et al., 2018, 2019). A significant number of pregnancies are affected by altered metabolic conditions of the mother, which is reflected in an abnormal intrauterine environment for the foetus. Along with abnormal maternal conditions are pregestational maternal obesity (PGMO) (Villalobos-Labra et al., 2018a) and supraphysiological
maternal gestational weight gain (spGWG) (according to the Institute of Medicine (IOM) and National Research Council (NRC) guideline) (IOM/NRC, 2009; Pardo et al., 2015), or pathologies of pregnancy including GDM (Subiabre et al., 2017; Cabalín et al., 2019) and preeclampsia (Salsoso et al., 2015; Mate et al., 2012, 2018). These metabolic alterations show with reduced insulin sensitivity and abnormal cell signalling in the foetoplacental vasculature. Since maternal obesity is nowadays considered a risk for developing GDM (Chu et al., 2007; Ramoni et al., 2017), a condition referred as gestational diabesity (Cabalín et al., 2019; Pardo and Sobrevia, 2018), insulin resistance will be a significant condition in pregnancies where the mother shows with this metabolic alteration.

3. Insulin resistance in obesity and T2DM

3.1. Obesity

Obese patients show a body mass index (BMI) $\geq 30$ kg/m$^2$ (WHO, 2018). Obesity associated with insulin resistance resembling dysregulation in the insulin signalling in target tissues (Tangseefa et al., 2018; Villalobos-Labra et al., 2018b). Nevertheless, endothelial dysfunction, a condition defined as an imbalance between vasodilating and vasoconstricting substances produced by (or acting on) endothelial cells (Deanfield et al., 2005), also relates to an altered insulin vascular signalling in the foeto-placental vasculature. Since maternal obesity is nowadays considered a risk for developing GDM (Chu et al., 2007; Ramoni et al., 2017), a condition referred as gestational diabesity (Cabalín et al., 2019; Pardo and Sobrevia, 2018), insulin resistance will be a significant condition in pregnancies where the mother shows with this metabolic alteration.
leptin, adiponectin) which regulate the insulin sensitivity in target tissues (Hardy et al., 2012; Mc Ardle et al., 2013; Villalobos-Labra et al., 2018b). Adipokines-regulation of insulin sensitivity includes an increase in the endoplasmic reticulum (ER) stress, a condition also associated with vascular insulin resistance in obesity in most tissues (Flamment et al., 2012; Villalobos-Labra et al., 2017, 2018b).

A higher release of the pro-inflammatory cytokines tumour necrosis factor α (TNF-α) and interleukin 6 (IL-6) is related to endothelial insulin insensitivity via a chronic inflammatory state leading to vascular insulin resistance (Engin, 2017). It is reported that the generation of pro-inflammatory mediators is more elevated in obese compared with non-obese subjects. Pathogenic activated macrophages (M1) from obese subjects show a higher release of TNF-α and IL-6 compared with M1 from non-obese subjects (Chawla et al., 2011). Interestingly, the adipose tissue in non-obese individuals also contains macrophages with an anti-inflammatory effect and capacity to releasing interleukin 10 (IL-10), called M2 macrophages. The M2 macrophages are proposed to be involved in enhancing the sensitivity to insulin by the adipose tissue (Lauterbach and Wunderlich, 2017; Russo and Lumeng, 2018). Since a correlation between higher M2 macrophages markers and increased release of pro-inflammatory cytokines is reported in patients with obesity, it is likely that the adipose tissue may also release molecules or react to molecules that protect against inflammation (Russo and Lumeng, 2018).

It is also reported that patients with obesity show increased plasma level of adenosine (Badillo et al., 2017; Cabalín et al., 2019; Johnston-Cox et al., 2012; Kaartinen et al., 1991; Pardo et al., 2017; Wojcik et al., 2014), a molecule that acts as anti-inflammatory via activation of A2B adenosine receptors (A2B AR) in most cell types and tissues (Antonioli et al., 2015; Cabalín et al., 2019; Pantham et al., 2015), including the foetoplacental endothelium (Cabalín et al., 2019). Adenosine causes an increase in the release of IL-10 requiring A2B AR activation in mouse macrophages (Nemeth et al., 2005) and in Escherichia coli-challenged mice macrophages (Čekša et al., 2007). Activation of A2B AR also caused an increase in the expression of arginase-1, tissue inhibitor of matrix metalloproteinase-1, and the macrophage galactose-type C-type lectin-1 leading to lower inflammation (Čekša et al., 2007). High plasma concentration of adenosine is found in inflammation (Cabalín et al., 2019; Fredholm, 2007; Sobrevia and Fredholm, 2017); thus, A2B AR activation-dependent reduced inflammation may result from lower generation of TNF-α, IL-6, and leptin, and higher production of IL-10 and adiponectin (for a review see Cabalín et al., 2019).

Other studies show increased free fatty acid (FFA) release in obesity associated with a higher generation of reactive oxygen species (ROS) by mitochondria which together with a lower eNOS activity in the endothelium results in endothelial dysfunction (Gremmels et al., 2015). However, nothing is reported regarding a role for FFA and the insulin response in the vasculature. Nevertheless, hyperglycaemia-increased ROS generation resulted in damaged endothelial cells (Li et al., 2017). Resistin has also been referred to as a pro-inflammatory adipokine which in human is mainly produced by the macrophages (Huang and Yang, 2016). Resistin associated with ER stress resulting in reduced insulin signalling in endothelial cells (Lou et al., 2018). Also, a reduced insulin-dilation mediated by resistin was reported in aortic and mesenteric segments in mouse (Gentile et al., 2008). The latter report addressed the possibility that resistin will reduced insulin sensitivity by lowering IRS-1/P3K/Akt/eNOS signalling. Thus, exacerbate adipose tissue during obesity produces endothelial dysfunction resulting in vascular insulin resistance.

Endothelial dysfunction associated with an altered generation of NO (Deanfield et al., 2005). Insulin causes vasodilation through the production of NO from the vascular endothelium (Sáez et al., 2019; Silva et al., 2017; Subiaire et al., 2019). Thus, insulin resistance will result in blunted endothelium-derived NO-dependent vascular response to this hormone. Obesity also associates with minor activation of Akt in endothelial cells, thus supporting the possibility that vascular endothelium is even less responsive to insulin in subjects with obesity. Several studies show low responsiveness of the endothelium to insulin and even a contractile effect of insulin in patients with obesity (Cardillo et al., 1999, 2004; Rocha et al., 2014). Nevertheless, this effect appears before the clinical manifestations of insulin resistance or diabetes mellitus in these patients. In primary cultures of HUVECs from women with normal pre-pregnancy BMI (20–24.9 kg/m²) that show rates of gestational weight gain (GWG) beyond the recommended range (i.e. spGWG) ending the pregnancy with obesity, insulin did not increase the NO generation (Pardo et al., 2015). Since ET-1 has a more substantial vasoconstrictor action in overweight or obese compared with lean subjects (Rocha et al., 2014), a delicate balance between NO and ET-1 on the vascular function is required (Paradis and Zhang, 2013).

Other studies report that HUVECs from women with PGMO showed ER stress, where the expression and activity of the ER stress sensors protein kinase RNA-like endoplasmic reticulum kinase, inositol-requiring enzyme 1α, and activating transcription factor 6 are increased (Villalobos-Labra et al., 2018a). PGMO-associated lower NO synthesis in HUVECs may also result from ER stress (Villalobos-Labra et al., 2017, 2018a). Other mechanisms involved in the foetoplacental vascular dysfunction in PGMO include (i) inhibition of IRS1/2 due to IR-A/p44/p42MAPK/S6K1 signalling, higher circulating levels of leptin and TNFα-dependent increased activity of c-Jun N-terminal kinase 1 (JNK1) (Villalobos-Labra et al., 2017), (ii) inhibition of IRS1/2 due to reduced inhibitory action mediated by adenosine monophosphate protein kinase (AMPK) on the mammalian target of rapamycin/p70 S6 kinase 1 (mTOR/S6K1) signalling as a result of lower circulating levels of adiponectin (Kim et al., 2015; Tzatsos, 2009), (iii) reduced activation of eNOS due to lower activator phosphorylation of serine 1177 or increased inhibitory phosphorylation of threonine 495, (iv) higher arginase activity, (v) increased L-arginine transport via hCAT-1 (and perhaps hCAT-2B) (Villalobos-Labra et al., 2018a).

3.2. T2DM

The worldwide number of individuals with diabetes mellitus is increasing from the last decade (ADA, 2018; WHO, 2018). This disease characterises by increased concentration of D-glucose in the plasma due to deficient uptake and abnormal metabolism by most (if not all) cells in the human body. Impaired intracellular homeostasis of D-glucose and other nutrients results in T2DM (~90% of individuals with diabetes mellitus) or GDM (~6–20% of pregnant women) (ADA, 2018; Egan et al., 2017; Melchoir, 2017; Subiabre et al., 2018; WHO, 2018). Patients with T2DM may show lower reactivity (or sensitivity) to insulin, abnormal insulin triggered cell signalling, and altered insulin receptors expression in target organs (WHO, 2018). T2DM was previously referred to as non-insulin–dependent diabetes mellitus, or adult-onset diabetes mellitus, and more recently it is a disease also diagnosed in children (WHO, 2018). It is now more evident that T2DM-associated altered signalling to insulin are worsened when patients are with overweight or obese. Indeed, an association between T2DM with obesity, overweight, and hyperinsulinaemia in response to the insulin resistance is seen in this disease (D’Adamo and Caprio, 2011; Salunkhe et al., 2017; Melchoir, 2017; Subiabre et al., 2018; WHO, 2018). The latter decade an increase in the ratio of new onset of T2DM compared with obesity and a stronger correlation with family history of T2DM is raising worldwide (WHO, 2017, 2018). Interestingly, obese men show lower risk than obese women to develop T2DM (Wilding, 2014). Thus, a different hormonal profile between men and women could be a component preconditioning to T2DM. Other risk factors to develop T2DM include signs of insulin resistance including hypertension, dyslipidaemia, and polycystic ovary syndrome (ADA, 2018; WHO, 2018).

Several studies report potential mechanisms for impaired endothelial insulin response in diabetes mellitus. These mechanisms include (i) overexpression of IR-A leading to preferential activation of p44/42MAPK versus Akt (p44/42MAPK/Akt > 1, i.e. mitogenic >
metabolic phenotype) in HUVeCs from GDM (Westermeier et al., 2011, 2015), (ii) downregulated IR-A expression but increased IR-B expression was paralleled by lower activation of Akt resulting in p44/42\textsuperscript{mapk}/Akt > 1 in human placenta microvascular endothelial cells (hPMECs) from GDM (Salomón et al., 2012), (iii) inhibition of IRS1/2 due to IR-A/ p44/42\textsuperscript{mapk}/Shc signalling, higher circulating levels of leptin, and TNF\alpha-dependent increased activity of JNK1, (iv) inhibition of IRS1/2 due to a lower inhibitory action mediated by AMPK on mTOR/S6K1 signalling as a result of lower circulating levels of adiponectin (Kim et al., 2015; Tzatsos, 2009), (v) higher inhibition of PI3K due to activation of the p85α regulatory subunit of PI3K (Hansen et al., 2001), (vi) increased expression of total eNOS and activator phosphorylation of serine 1177 (Ser1177) (Subiabre et al., 2017), and (ix) increased L-arginine transport via hCAT-1 (Subiabre et al., 2018).

In an interesting study, cultured freshly isolated forearm vein endothelial cells from patients with T2DM were challenged for 30 min with insulin (Tabit et al., 2013). Insulin caused higher phosphorylation of eNOS at Ser1177 in cells from healthy subjects but reduced Ser1177 phosphorylation in cells from subjects with T2DM. However, basal Ser1177 phosphorylation in cells from T2DM was higher (∼1.7 fold) than in non-diabetic subjects. The basal increased eNOS activity seen in cells from T2DM subjects was reversed by LY379196, a pharmacological inhibitor of protein kinase C β (PKC β), suggesting the involvement of this kinase in this phenomenon. Interestingly, inhibition of PKC β improved the response to insulin reversing the elevated Ser1177 phosphorylation of eNOS. Thus, this protein kinase may act blocking the response to insulin increasing insulin resistance in T2DM. Interestingly, more of the changes detected in endothelial cells subjects with T2DM and GDM are reproduced in HUVeCs isolated from normal pregnancies but incubated with 25 mmol/L D-glucose for periods up to 24 h in vitro (Montecinos et al., 2000; Sobrevia et al., 1998).

The findings described in adult peripheral endothelial cells (Tabit et al., 2013) are similar to those described in HUVeCs from GDM pregnancies, where PKC was also involved in the basal increase in eNOS activity (Vásquez et al., 2004). Moreover, incubation of HUVeCs with 25 mmol/L D-glucose increased Ser1177 phosphorylation and NO activity, a phenomenon that required PKC activity (Montecinos et al., 2000). Interestingly, insulin restored the GDM-elevated L-arginine transport and NO synthesis in HUVeCs, but this effect was absent when cells from GDM pregnancies were exposed to 25 mmol/L D-glucose (Sobrevia et al., 1998). Thus, insulin resistance in endothelial cells from subjects with diabetes mellitus (T2DM or GDM) may result from these diseases-associated hyperglycaemia-increased PKC (likely PKC β) activity (Montecinos et al., 2000; Sobrevia et al., 1998; Tabit et al., 2013; Vásquez et al., 2004). The fact that altered signalling mechanisms leading to endothelial dysfunction are also seen in cells exposed to hyperglycaemia suggests that elevated D-glucose level in diabetes mellitus may be responsible at least in part of the altered vascular responsiveness to insulin. However, hyperglycaemia may also not be a factor leading to altered endothelial (and vascular) function. The glycaemia of pregnant women with GDM pregnancies treated with diet or insulin was normal as it was in the umbilical vein blood (Subiabre et al., 2017; Villalobos-Labra et al., 2018a). However, HUVeCs and hPMECs from GDM pregnancies with normal maternal and umbilical blood levels of D-glucose showed activation of L-arginine/NO signalling pathway (Subiabre et al., 2017). Thus, plasma D-glucose level may not be the only factor leading to endothelial dysfunction in the foeto-placental vasculature (Sobrevia et al., 2015; Subiabre et al., 2017, 2018, 2019).

4. Vascular alterations in diabesity

Diabesity is the term used to define the result of a combination of factors determining metabolic disorders with a state of insulin resistance (Kalra, 2013; Potenza et al., 2017; Ziv and Sharif, 1995). Patients that are obese and show with T2DM present diabesity (Kalra, 2013; Ziv and Sharif, 1995). Diabesity subjects show a pattern of clinical alterations that are unique for this metabolic condition and do not precisely correspond to clinical modifications seen in individuals affected by only obesity or T2DM. However, patients considered as part of the diabesity group present with common characteristics described for obesity or T2DM including endothelial and vascular dysfunction in response to vasoactive endogenous molecules such as insulin, adenosine, or ET-1 (Cabalín et al., 2019; Campia et al., 2014; Pardo et al., 2017; Sáez et al., 2019; Silva et al., 2017, 2019) (Fig. 3).

4.1. Diabesity-associated vascular alterations

Mild hypoglycaemia is seen in patients with diabesity that are treated for T2DM, a phenomenon that results in endothelial dysfunction and increased pro-inflammatory response (Joy et al., 2016). The latter study compared obese patients with T2DM (diabesity) versus non-obese healthy controls but did not distinguish patients showing only T2DM (i.e., non-obese T2DM patients). Their results suggest that diabesity will result in endothelial dysregulation due to reduced plasma levels of vascular cell adhesion molecule 1 and tissue plasminogen activator. Interestingly, unaltered ET-1, intercellular adhesion molecule 1, P-selectin, E-selectin, plasminogen activator inhibitor-1 (PAI-1), and TNF-α plasma levels were reported. Since activation of TNF-α is a condition associated with insulin resistance (Villalobos-Labra et al., 2017) and ET-1 is higher in patients with obesity or T2DM, it is likely that diabesity may reflect a different combination of these factors (and perhaps additional factors) leading to insulin resistance compared with T2DM or obesity. An earlier study suggested the role of ET-1 and NO in the vascular reactivity in patients with diabesity (Mather et al., 2002). The results of that study support the possibility that ET-1 contributed to endothelial dysfunction and the regulation of vascular tone in human obesity as well as in patients with diabesity but did not address whether these alterations were seen in patients with T2DM alone. Thus, it is still undefined whether ET-1– and NO-associated vascular dysfunction in diabesity was due to diabesity, obesity, or T2DM.

One of the roles of insulin is maintaining a balance between NO and ET-1 in several tissues including the vascular endothelium (Mahmoud et al., 2016; Reynolds et al., 1985; Villalobos-Labra et al., 2017). In
these studies, T2DM patients with BMI varying from overweight to obesity, i.e., some of them overweight with T2DM and others with diabesity, hyperinsulinemia increases ET-1 release reducing the eNOS activation and reducing the dilation in human skeletal muscle arterioles. Thus, a vasoconstrictive pathway that impaired the arteriolar vasodilation in likely in these subjects. Unfortunately, patients were all considered in the same group with T2DM even when some of them were overweight and others with obesity. Thus, not a proper conclusion regarding the potential effects of diabesity on endothelial dysfunction due to ET-1 and NO physiological balance can be made from this study.

The insulin-stimulated leg blood flow was estimated in subjects with diabesity and in individuals that were not with T2DM but in a mix being overweight and obese (BMI 33.4–36.2 kg/m²) (Reynolds et al., 1985). The results show that vascular response to insulin is bluntly likely due to reduced eNOS activation, whether the altered vascular response resulted from an imbalance between ET-1 expression and activity is unclear since only the mRNA but not the protein level of ET-1 was higher in patients with diabesity compared with normal healthy lean subjects (Reynolds et al., 1985). Even when similar alterations were indicated for the referred non-obese patients with T2DM, this conclusion is questionable since the study included a mix of patients with overweight and obesity. Thus, lacking from proper controls for obesity or T2DM alone, an outcome regarding diabesity vascular effects cannot be given.

The foetoplacental vasculature reacts to insulin with vasodilation, a response that is impaired in PGMO (Villalobos-Labra et al., 2018a, 2018b) and women with spGWG (Pardo et al., 2015). The mechanisms involved in the reduced response to insulin include lower activation of IRS1, a key mediator of the IR-B-triggered response in human endothelium (Westermeier et al., 2016; Yoneyama et al., 2015). Also, PGMO and spGWG resulted in reduced NO synthesis in HUVECs, a phenomenon that perhaps was due to reduced activation of IRS1-dependent Akt activity resulting in lower activation of mTOR limiting the vascular insulin signalling in endothelial cells (Villalobos-Labra et al., 2017). Interestingly, periumbilical subcutaneous adipose tissue and small subcutaneous arterioles (~220 μm diameter) from obese patients showed defective insulin signalling in response to insulin compared with lean subjects (Georgescu et al., 2011). Patients that were with diabesity showed reduced eNOS protein expression and activity, endothelium-dependent and insulin-induced vasodilatation, IRS1/2, PI3K, and Akt protein expression and activity compared with patients with obesity or lean subjects. Thus, additional elements may play a role in causing endothelial and vascular dysfunction in patients that are with diabesity compared with those with obesity only.

4.2. Gestational diabesity?

Pregnancy is a physiological state where the mother is subjected to extreme stress altering almost all the systemic adaptive mechanisms. Physiological insulin resistance results in response to elevate D-glucose plasma levels due to the requirements of the growing foetus. However, when the women show with pre-pregnancy obesity or overweight the physiological conditions are altered, and the foetus growth and development is abnormal (Pardo et al., 2017, 2018; Villalobos-Labra et al., 2017, 2018a,b). Interestingly, most of the studies addressing GDM effects in the foetoplacental vasculature included women with PGMO or overweight instead of referring to GDM in women with normal pre-pregnancy weight. An increase in GDM and T2DM incidence in parallel with obesity is seen worldwide in the last decade (ADA, 2018). However, no studies address whether obesity, without T2DM (or T1DM), in women that become pregnant and develop GDM could worsen the maternal, foetal, and newborn outcome caused by PGMO, spGWG, or GDM by itself (Cabalín et al., 2019). GDM plus obesity (or overweight) may be a different metabolic condition compared with diabesity since it occurs in a different physiological context (i.e. pregnancy in this case) and involves the development and growth of the foetus. This condition, i.e. GDM in obese pregnant women, has been introduced as gestational diabesity (Cabalín et al., 2019; Pardo and Sobrevia, 2018; Sáez et al., 2019). Unfortunately, whether maternal obesity in pregnancy with overt T2DM and T1DM may result in a different kind of diabesity when the mother develops GDM is not documented. Therefore, the immediate, short, middle and long consequences of a potential metabolic state of gestational diabesity are still unknown.

5. Conclusions

The incidence of T2DM and obesity worldwide is rapidly increasing and the concurrence of both diseases, i.e. diabesity, results in metabolic alterations that are different from those reported in subjects with obesity or T2DM by itself. The majority of the patients with T2DM are obese, and a common characteristic in these subjects is the abnormal response of the vasculature to insulin. Similar mechanisms are seen regarding vascular insulin resistance in obesity and T2DM, and even in GDM; nevertheless, the majority of the patients with T2DM or mothers with GDM show excess weight (overweight or obesity) thus being difficult to discriminate the direct effect on the vasculature of each of these pathologies itself (Fig. 4).

Most studies reporting data in women with GDM did not separate these patients in obese or overweight in pregnancy or before pregnancy (i.e., PGMO or pre-pregnancy maternal overweight) versus women with GDM that were with pre-pregnancy normal weight or showed physiological GWG. Since obesity, PGMO and GDM result in different physiological alterations in the foetoplacental vasculature (Pardo et al., 2015; Westermeier et al., 2015; Villalobos-Labra et al., 2018a), it is likely that cellular mechanisms leading to insulin resistance in the foetoplacental vasculature in these altered metabolic conditions are different. It is now more evident that patients with obesity that develop T2DM, i.e. diabesity, show worsened vascular reactivity to insulin compared with obese or T2DM patients. Women with gestational diabesity will show vascular insulin signalling alterations that may or may not be similar to those seen in GDM pregnancies (Cabalín et al., 2019; Pardo and Sobrevia, 2018). The normalisation of obesity- and GDM-associated foetoplacental endothelial dysfunction by insulin may or may not result from similar mechanisms in gestational diabesity. Several studies show an association between the detrimental effects of obesity in pregnancy and GDM with sexual dimorphism. To date, female foetus to normal pregnancies associated with lower risk of
developing early maternal pregnancy insulin resistance (estimated by HOMA-IR assay) compared with the male foetus (Walsh et al., 2015). Another study shows that obese women with a male foetus had highest oxidative and nitrite stress, a phenomenon contributing to maternal obesity-associated adverse outcomes with a male foetus (Evans and Myatt, 2017). Foetal sex is also determinant in the risk to develop T2DM and GDM (Retnakaran and Shah, 2015). A higher risk of developing GDM in the first and second pregnancy is seen with a male foetus, women with GDM in the first pregnancy show a higher risk of developing T2DM with female foetus, and women carrying a female foetus in a healthy first pregnancy but having a male in the second pregnancy predicted a higher risk of GDM (Retnakaran and Shah, 2015). Thus, sexual dimorphism is involved in adverse outcomes for the mother and newborn to pregnancies with maternal obesity or GDM. A different result related to the foetus gender in diabesity or gestational diabesity is expected but not yet described (Pardo and Sobrevia, 2018; Cabalín et al., 2019). As a general conclusion, we emphasis the fact that further studies are needed to address diabesity and to get a better understanding of the several physiological consequences of this abnormal metabolic condition in these patients.

Conflicts of interest

The authors confirm that there are no conflicts of interest.

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