Microvascular dysfunction: causative role in the association between hypertension, insulin resistance and the metabolic syndrome?


*Department of Internal Medicine, VU Medical Center, PO Box 7057, 1007 MB Amsterdam, The Netherlands, †Laboratory for Physiology, Institute for Cardiovascular Research, VU Medical Center, Amsterdam, The Netherlands, and ‡Department of Internal Medicine, Academic Hospital Maastricht, PO Box 5800, 6202 AZ Maastricht, The Netherlands

Abstract

The metabolic syndrome defines a clustering of metabolic risk factors that confers an increased risk for type 2 diabetes and cardiovascular disease. The metabolic syndrome seems to have multiple etiological factors and microvascular dysfunction may be one potential factor explaining the clustering of multiple metabolic risk factors including hypertension, obesity, insulin resistance and glucose intolerance. Microvascular dysfunction may increase not only peripheral vascular resistance and blood pressure, but may also decrease insulin-mediated glucose uptake in muscle. The present article summarizes some of the data concerning the role of microvascular dysfunction in the metabolic syndrome.

†To whom correspondence should be addressed (email e.serne@vumc.nl).
Introduction

The metabolic syndrome defines a clustering of metabolic risk factors that confers an increased risk for type 2 diabetes and cardiovascular disease [1]. Obesity and a central body fat distribution, hypertension, insulin resistance, glucose intolerance, dyslipidaemia and proinflammatory and prothrombotic factors are all part of the metabolic syndrome. Previously, a large amount of research has been aimed at elucidating the pathophysiology underlying this clustering of risk factors, since a better understanding may lead to new therapeutic approaches that specifically target underlying causes of the metabolic syndrome.

Recently it has become clear that microvascular dysfunction, by affecting both pressure and flow patterns, may have consequences not only for peripheral vascular resistance, but also for insulin-mediated changes in muscle perfusion and glucose metabolism, shedding new light on the association between hypertension, obesity and impaired insulin-mediated glucose disposal [2–4]. An important consequence of this concept is that any condition that impairs microvascular function will predispose to both insulin resistance and hypertension. The present article examines some of the data concerning the role of microvascular dysfunction as an explanation for the association between hypertension, obesity and impaired insulin-mediated glucose disposal.

Description of the microcirculation

The microcirculation is widely taken to encompass vessels < 150 μm in diameter. It therefore includes arterioles, capillaries and venules. Nowadays, a definition based on arterial vessel physiology rather than diameter or structure has been proposed, depending on the response of the isolated vessel to increased internal pressure [3]. By this definition, all those arterial vessels that respond to increasing pressure by a myogenic reduction in lumen diameter would be included in the microcirculation. Such a definition would include the smallest arteries and arterioles in the microcirculation in addition to capillaries and venules. Small arterial and arteriolar components should, therefore, be considered a continuum rather than distinct sites of resistance control.

A primary function of the microcirculation is to optimize nutrient and oxygen supply within the tissue in response to variations in demand. A second important function is to avoid large fluctuations in hydrostatic pressure at the level of the capillaries causing disturbances in capillary exchange. Finally, it is at the level of the microcirculation that a substantial proportion of the drop in hydrostatic pressure occurs. The microcirculation is therefore extremely important in determining the overall peripheral resistance.
**Microvascular function is impaired in hypertension and obesity**

In hypertension, the structure and function of the microcirculation are altered in at least three ways [3,5]. First, the mechanisms regulating vasomotor tone are abnormal, leading to enhanced vasoconstriction or reduced vasodilator responses. Secondly, there are anatomical alterations in the structure of individual precapillary resistance vessels, such as an increase in their wall-to-lumen ratio. Finally, there are changes at the level of the microvascular network involving a reduction in the number of arterioles or capillaries within vascular beds of various tissues (e.g. muscle and skin), so called vascular rarefaction [3,5,6]. In obese individuals similar defects in the microcirculation can be demonstrated [7]. Enhanced vasoconstriction and reduced vasodilator responses can be demonstrated in the microcirculation of obese subjects [7]. Rarefaction of arterioles and capillaries within vascular beds of various tissues (e.g. muscle and skin) can also be demonstrated [7]. In addition, measures of obesity in healthy individuals are strongly related to skin microvascular function [2,7].

Taken together, microvascular dysfunction in different tissues has been established in both hypertension and obesity.

**Hypertension as a result of microvascular dysfunction**

In most forms of experimental and clinical hypertension, cardiac output is close to normal and the peripheral vascular resistance is increased in proportion to the increase in blood pressure [3]. There is general agreement that there is relatively little pressure loss within the large conduit arteries and that the drop in pressure occurs predominantly in vessels ranging from 10 to 300 μm in diameter. The increase in total peripheral vascular resistance, is likely to reflect changes in these vessels. Whereas it has been known for many years that increased wall-to-lumen ratio and microvascular rarefaction can be secondary to sustained elevation of blood pressure [3], there is also evidence that abnormalities in the microcirculation precede and thus may be a causal component of high blood pressure. Microvascular rarefaction similar in magnitude to the rarefaction observed in patients with established hypertension can already be demonstrated in subjects with mild intermittent hypertension, and in normotensive subjects with a genetic predisposition to high blood pressure [3,5,6]. In several tissues capillary density has been found to correlate inversely with peripheral vascular resistance and blood pressure in hypertensive, normotensive lean and normotensive obese subjects [2,6,7]. Moreover, in hypertensive subjects, capillary rarefaction in muscle has been shown to predict the increase in mean arterial pressure over two decades [8]. More recently, a smaller retinal arteriolar diameter has been shown to predict the occurrence and development of hypertension in a
prospective, population-based study of normotensive middle-aged persons [9]. In addition, mathematical modelling of in vivo microvascular networks predicts an exponential relationship between capillary and arteriolar number and vascular resistance. Total vessel rarefaction up to 42% (within the range observed in hypertensive humans) can increase tissue vascular resistance by 21%. Thus it seems likely that microvascular abnormalities can both result from and contribute to hypertension, and a ‘vicious cycle’ may exist in which the microcirculation maintains or even amplifies an initial increase in blood pressure. It has been demonstrated that an initial small increase in pressure can lead to larger structural increases in pressure and flow resistance by a mechanism involving the tendency of vessels to reduce their luminal diameter in response to increased intraluminal pressure [3]. This argument could be taken a step further to suggest that microvascular abnormalities causing an increase in peripheral resistance might initiate the pathogenic sequence in primary hypertension. However, according to the Borst-Guyton concept, chronic hypertension can only occur if renal function is abnormal with a shift in the renal pressure–natriuresis relationship. In the absence of the latter, increased peripheral resistance only temporarily raises blood pressure, to be followed by an increase in renal sodium excretion restoring blood pressure towards normal. Importantly, therefore, subtle renal microvascular disease [10] as well as a reduced number of nephrons [11] may reconcile the Borst-Guyton concept with the putative role of vessel rarefaction in the etiology of high blood pressure. This may also explain the relationship between salt sensitivity of blood pressure, a characteristic of hypertension associated with the metabolic syndrome and insulin resistance [12].

It is important to realize that a decreased capillary density also affects the spatial pattern of flow in the microvascular bed, causing a non-uniform distribution of blood flow among exchange vessels. This non-uniform distribution of flow among vessels, which can be defined as some vessels receiving more and some less of their appropriate fraction of total flow, has been invoked to explain phenomena such as flow-limited muscular performance [13] and sub-optimal capillary transport of small solutes [14]. In addition, it may contribute to various kinds of end-organ damage (e.g. retinopathy, lacunar stroke, microalbuminuria and heart failure) [3].

In summary, microvascular dysfunction, in particular rarefaction, by affecting both pressure and flow patterns, may have consequences not only for peripheral vascular resistance and blood pressure, but also for muscle perfusion and metabolism.

**Insulin resistance as a result of microvascular dysfunction**

Insulin resistance is typically defined as decreased sensitivity and/or responsiveness to metabolic actions of insulin that promote glucose disposal. A major action of insulin in muscle tissue involves translocation of glucose
transporters to the plasma membrane and activation of downstream pathways of glucose metabolism [15]. The glucose transporter protein is considered to be rate-limiting for insulin-stimulated glucose uptake in the muscle [15]. However, before insulin interacts with the receptor on the plasma membrane, insulin and glucose must be delivered to the muscle cells at normal levels and at the correct time. Recently, there has been a surge of interest in these pre-cellular steps, in particular with regard to the possible contribution of insulin-mediated changes in muscle blood flow to insulin-mediated glucose uptake.

Insulin increases total blood flow and blood volume in skeletal muscle [4]. Principally because the ability of insulin to dilate skeletal muscle vasculature is impaired in a wide range of insulin-resistant states (e.g. hypertension, obesity and type 2 diabetes), it has been hypothesized that insulin’s vasodilatory and metabolic actions (i.e. glucose disposal) are functionally coupled [4,16,17]. However, despite the compelling nature of these findings, the concept that insulin might control its own access and that of other substances, particularly glucose, has been vigorously challenged. By approaching the experiments differently, in particular with lower doses of insulin and shorter time courses, it was shown that insulin-mediated changes in total blood flow appear to have time kinetics and a dose dependence on insulin different from those for the effect on glucose uptake. In addition, studies in which glucose uptake has been measured during hyperinsulinaemia and manipulation of total limb blood flow with different vasodilators have shown that total limb blood flow could be increased in either normal or insulin-resistant individuals, yet there was no increase in insulin-mediated glucose uptake [4]. The discrepancy in these findings has been ascribed to the fact that various vasoactive agents may change total flow but have distinct effects on the microcirculation and on the distribution of blood flow in nutritive compared with non-nutritive vessels. Clark et al. have introduced the concept that distribution of blood flow in nutritive compared with non-nutritive vessels, independent of total muscle flow, may affect insulin-mediated glucose uptake [4]. Using studies in rats, applying different approaches to measure capillary recruitment (1-methylxanthine metabolism) and microvascular perfusion [CEU (contrast-enhanced ultrasound) and laser Doppler flowmetry], it could be demonstrated that insulin mediates changes in muscle microvascular perfusion consistent with capillary recruitment [4]. This capillary recruitment relates to changes in skeletal muscle glucose uptake independently of changes in total blood flow, requires lower insulin concentrations, and precedes muscle glucose disposal [4,17]. This has led to the hypothesis that insulin, possibly by reducing precapillary arteriolar tone and/or altering arteriolar vasomotion, redirects blood flow from non-nutritive vessels to nutritive capillary beds, resulting in an increased and more homogeneous overall capillary perfusion termed ‘functional capillary recruitment’. The latter would enhance the access of insulin and glucose to a greater mass of muscle for metabolism. Consistent with such a mechanism in
humans, insulin increases microvascular blood volume as measured with CEU or positron emission tomography and enhances the distribution volume of glucose in human muscle [3,17]. We have shown, by directly visualizing capillaries in human skin, that systemic hyperinsulinaemia is capable of increasing the number of perfused capillaries [7,16]. This insulin-dependent capillary recruitment is impaired in obese insulin-resistant subjects [7,18]. Moreover, it is associated with the number of capillaries recruited during post-occlusive reactive hyperaemia without insulin infusion, a measure of capillary recruitment which has been shown to be related to insulin-mediated whole body glucose uptake [2,6,7] and to be decreased in insulin-resistant hypertensive and obese subjects [6,7]. Making use of iontophoresis and laser Doppler flowmetry, we could also demonstrate that locally applied insulin induced microvascular vasodilation in human skin, independently of insulin’s systemic effects [16]. Furthermore, we could demonstrate that systemic hyperinsulinaemia influences microvascular vasomotion in human skin [16] and muscle [19].

Vasomotion, the rhythmic fluctuations of microvascular blood flow, may be an important determinant of the spatial and temporal heterogeneity of microvascular perfusion and, therefore, of the number of perfused capillaries [19]. The origin and control of microvascular vasomotion is still a matter of debate. A central neurogenic regulatory mechanism is suggested by synchronicity on contralateral limbs and by the suppressive effect of central sympathectomy. However, local administration of vasoactive substances such as acetylcholine and sodium nitroprusside directly influences vasomotion. Furthermore, vasomotion has been shown in isolated small arteries, indicating a local regulatory mechanism. In view of these considerations, it can be suggested that vasomotion is regulated by both local vasoactive substances and influences of the central nervous system. The contribution of different regulatory mechanisms can be investigated by analysing the contribution of different frequency intervals to the variability of the laser Doppler signal. Our data suggest that an insulin-mediated effect on microvascular vasomotion occurs by increasing endothelial and neurogenic activity [16,19].

Further insight into the complex relationships among vasodilation, blood flow velocity and capillary recruitment was gained through measurement of the PS (capillary permeability-surface area product) for glucose and insulin. The PS for a substance describes its capacity to reach the interstitial fluid. This depends on the permeability and the capillary surface area, which in turn depends on the extent of capillary recruitment.

A recent investigation employing direct measurements of muscle capillary permeability showed that PS for glucose increased after an oral glucose load, and a further increase was demonstrated during an insulin infusion [20]. The increase of PS was exerted without any concomitant change in total blood flow. It was concluded that the insulin-mediated increase in PS seen after oral glucose is important for the glucose uptake rate in normal muscle [20].
Interestingly, PS for glucose is subnormal under steady-state insulin clamp conditions in insulin-resistant type 2 diabetic subjects [20]. Moreover, a close and positive correlation was demonstrated between the rate of muscle glucose uptake and PS for glucose. A stimulated uptake of glucose and insulin in the absence of an increased PS would, hypothetically, lead to depletion of these substances and a lowered interstitial concentration. Importantly, at steady state levels, the interstitial muscle insulin and glucose concentrations nevertheless were normal in the type 2 diabetes group. The concomitant cellular insulin resistance leading to a subnormal glucose uptake rate may balance the low transcapillary transport rate of glucose and insulin so that the interstitial fluid concentrations stay normal [20]. The importance of the perturbed capillary recruitment for the reduction in glucose uptake is evident, however, because a normal increase in PS in type 2 diabetes muscle would lead to supernormal interstitial concentrations [20].

Another aspect of insulin resistance is a delay in insulin action [21]. Previously it has been reported that the time of onset of insulin action is delayed in insulin-resistant obese, type 2 diabetic and hypertensive subjects. A delayed transcapillary insulin transport in insulin-resistant states has been reported from \textit{in vitro} studies and some human \textit{in vivo} studies. Moreover, in obese subjects, this delay in insulin action was accompanied by a slow delivery of insulin to the muscle interstitial fluid during insulin/glucose infusion [21].

These data illustrate the importance of the microcirculation in regulating nutrient and hormone access to muscle, and raise the possibility that any impairment in capillary recruitment may cause an impairment in glucose uptake by muscle.

**Mechanisms involved in impairment of insulin-mediated capillary recruitment**

**Vascular insulin resistance**

Insulin-stimulated glucose uptake in skeletal muscle and adipose tissue is mediated by translocation of the insulin-responsive glucose transporter GLUT4 to the cell surface. This requires PI3K (phosphatidylinositol 3-kinase)-dependent signalling pathways that involve the insulin receptor, IRS-1 (insulin receptor substrate-1), PI3K, PDK1 (phosphoinositide-dependent kinase 1) and Akt (protein kinase B) [22]. Ras/MAPK (mitogen-activated protein kinase) pathways do not contribute significantly to insulin-stimulated translocation of GLUT4, but are important for insulin-mediated regulation of growth and mitogenesis [23,24]. Interestingly, the vascular actions of insulin that stimulate the production of NO (nitric oxide) require PI3K-dependent insulin-signalling pathways that bear striking similarities to metabolic insulin-signalling pathways (Figure 2). Moreover, the MAPK branch of insulin signalling controls secretion of ET-1 (endothelin-1), a strong vasoconstrictor,
by the endothelium [22–24]. Insulin has therefore opposing haemodynamic actions on vessels. In vessels from healthy rats, insulin has no net effect on vessel diameter, because of a balance between the stimulation of two pathways, NO-mediated vasodilation and ET-1-mediated vasoconstriction. Insulin stimulates activation of endothelial NO synthase: the signalling pathway is through IRS-1, PI3K and Akt [22]. However, if this pathway is inhibited, the arteriole constricts, a response mediated by ET-1 through the Ras/MAPK and ERK1/2 (extracellular signal-related kinase-1/2) pathway [24]. These observations imply a dual insulin signalling mechanism in vessels, one pathway stimulating the synthesis of NO, the other stimulating ET-1 release. In obese rats, these signalling pathways are selectively impaired: insulin-mediated activation of the ET-1 pathway is impaired, but insulin-mediated activation of ERK1/2 is intact [25]. In line with this evidence, we have recently found insulin-induced, ET-1-dependent vasoconstriction in skeletal muscle arterioles of obese rats (E.C. Eringa, C.D.A. Stehouwer, M.H. Roos, N. Westerhof and P. Sipkema, unpublished work). In addition, insulin resistance in spontaneously hypertensive rats is associated with endothelial dysfunction characterized by an imbalance between NO and ET-1 production [26]. Moreover, obese, hypertensive individuals show an insulin-induced vasoconstriction and increased ET-1-dependent vasoconstrictor tone and decreased NO-dependent vasodilator tone at the level of the resistance arteries [27]. Thus, shared insulin-signalling pathways in metabolic and vascular target tissues with complementary functions may provide a mechanism to couple the regulation of glucose and haemodynamic homoeostasis. The net haemodynamic action of insulin is dependent on a balance between its vasodilator and vasoconstrictor effects. An imbalance between NO and ET-1 production may explain insulin resistance-related hypertension.

**Obesity-related endocrine signalling**

The close association between measures of adiposity and microvascular function necessitates communicative pathways between adipose tissue and the microvasculature. Adipose tissue and in particular visceral adipose tissue cells secrete a variety of bioactive substances called adipokines such as NEFAs (non-esterified fatty acids), adiponectin, leptin, resistin, angiotensinogen and TNF-α (tumour necrosis factor-α). In the next section we will focus on the role of NEFAs and TNF-α.

Using magnetic resonance spectroscopy, NEFA-induced insulin resistance in humans has been shown to result from a significant reduction in the intramyocellular glucose concentration, suggestive of glucose transport as the affected rate-limiting step [15]. The current hypothesis, supported by data from PKC-θ (protein kinase Cθ) knockout mice, proposes that fatty acids upon entering the muscle cell activate PKC-θ as either fatty acyl-CoA or diacylglycerol. The PKC-θ activates a serine kinase cascade leading to the phosphorylation...
and inactivation of IRS-1 by preventing its activation by tyrosine phosphorylation [23]. Since the technique of magnetic resonance spectroscopy only identifies a gradient from extracellular to intracellular glucose in muscle cells, it remains to be proven that the gradient did not occur between the plasma and interstitial glucose and thus reflects a rate limiting step of glucose delivery induced by fatty acids. Interestingly, studies suggest that glucose delivery contributes to sustaining the transmembrane glucose gradient and, therefore, is a determinant of glucose transport [28]. This would be consistent with the finding in rats that NEFA elevation concomitantly impairs insulin-mediated muscle capillary recruitment and glucose uptake [4,17]. In addition, we could demonstrate that in lean individuals, NEFA elevation induces skin microvascular dysfunction and reduces whole body glucose uptake, while in obese individuals NEFA lowering has the opposite effect (Figure 1) [18]. Moreover, changes in capillary recruitment statistically explained approx. 29% of the association between changes in NEFA levels and insulin-mediated glucose uptake. A defect involving fatty-acid-induced impaired insulin signalling through the same PKC-θ mechanism in endothelial cells, which in turn may negatively influence the balance between insulin-mediated vasodilation and vasoconstriction, may be responsible for the impaired capillary recruitment.

Figure 1. Capillary recruitment (%) before and during hyperinsulinaemia in obese women
Effects of NEFA lowering versus placebo (A). Capillary recruitment (%) before and during hyperinsulinaemia in lean women. Effects of NEFA elevation versus saline infusion (control) (B).
Increased production of the proinflammatory cytokine TNF-α is associated with obesity-related insulin resistance [29] as well as obesity-related hypertension [30]. In rats, TNF-α elevation concomitantly impairs insulin-mediated muscle capillary recruitment and glucose uptake [4,17]. In addition, in humans, circulating TNF-α levels are associated with reduced whole body glucose uptake and skin capillary recruitment [29]. In isolated skeletal muscle resistance arteries, we could demonstrate that TNF-α impairs the vasodilator effects but not the vasoconstrictor effects of insulin through activation of the intracellular enzyme JNK (c-Jun N-terminal kinase) and impairment of insulin-mediated activation of Akt [31]. This selective inhibition of the vasodilator effects of insulin results in insulin-mediated vasoconstriction in the presence of TNF-α. JNK has been shown to regulate whole-body insulin sensitivity as well as insulin-mediated cell signalling [32]. In conclusion, both NEFA and TNF-α are likely candidates to link visceral adipose tissue with defects in microvascular function, at least in part by influencing insulin signalling and thereby insulin’s vascular effects.

Vasocrine signalling

We have recently hypothesized an alternative communicative pathway [33]. Obese Zucker rats are characterized by a well-circumscribed depot of fat cells around the origin of the nutritive arteriole supplying the cremaster muscle whereas lean rats are not. Adipokines released by these fat cells may directly inhibit vasodilatory pathways distal in the arteriole and thereby cause loss of blood flow in the nutritive capillary network supplied by this arteriole (Figure 2).

![Figure 2. Mechanisms of insulin-mediated NO and ET-1 production leading to vasoconstriction and vasodilation respectively](image)

Adipokines secreted by (perivascular) adipocytes inhibit the PI3K pathway of insulin signalling. eNOS, endothelial nitric oxide synthase.
In this hypothesis, which remains to be tested, adipokines released from periarteriolar fat depots have a local rather than a systemic vasoregulatory effect, which we named ‘vasocrine’.

**Conclusion**

The metabolic syndrome defines a clustering of metabolic risk factors that confers an increased risk for type 2 diabetes and cardiovascular disease. The metabolic syndrome seems to have multiple etiological factors. A complex interaction between microvascular function, intracellular insulin signalling pathways and obesity-related endocrine signalling molecules may be one potential factor explaining the clustering of multiple metabolic risk factors such as hypertension, obesity, insulin resistance and glucose intolerance. A better understanding of the pathophysiology underlying the clustering of risk factors may lead to new therapeutic approaches that specifically target underlying causes of the metabolic syndrome. Microvascular dysfunction may play a central role by increasing not only peripheral vascular resistance and blood pressure, but also decreasing insulin-mediated glucose uptake in target cells (Figure 3).

![Diagram of microvascular dysfunction](image)

**Figure 3. Microvascular dysfunction: postulated causative role in the association between hypertension, insulin resistance and the metabolic syndrome**

Microvascular dysfunction of any cause may play a central role by increasing not only peripheral vascular resistance and blood pressure, but also decreasing insulin-mediated glucose uptake in target cells.
Summary

• The metabolic syndrome defines a clustering of metabolic risk factors that confers an increased risk for type 2 diabetes and cardiovascular disease.

• Microvascular dysfunction, may be one potential factor explaining part of this clustering of multiple metabolic risk factors including hypertension, obesity and insulin resistance.

• Microvascular abnormalities such as vascular rarefaction can cause an increase in peripheral resistance and might initiate the pathogenic sequence in hypertension.

• Insulin has direct effects (increasing glucose uptake in skeletal muscle) and substantial indirect effects (promoting glucose disposal by redistributing blood flow from non-nutritive to nutritive vessels). This cross-talk between metabolic and vascular tissues is important for coupling glucose homeostasis and (micro)vascular function.

• Shared insulin-signalling pathways in metabolic and vascular target tissues may provide a mechanism to couple the regulation of glucose and haemodynamic homeostasis. Metabolic insulin resistance is characterized by pathway-specific impairment in PI3K-dependent signalling, which in endothelium may cause imbalance between production of NO and secretion of ET-1, leading to decreased microvascular perfusion and an impairment in glucose uptake.

• NEFAs and proinflammatory cytokines including TNF-α may contribute to impairment of insulin’s metabolic and vascular actions by modulating insulin signalling and transcription.

References


© 2006 The Biochemical Society


