



Vascular Health Triad in Humans With Hypertension—Not the Usual Suspects

Sushant M. Ranadive^{1*}, Gabrielle A. Dillon^{2,3}, Sara E. Mascone¹ and Lacy M. Alexander^{2,3}

¹ Department of Kinesiology, University of Maryland, College Park, College Park, MD, United States, ² Department of Kinesiology, The Pennsylvania State University, University Park, PA, United States, ³ Center for Healthy Aging, The Pennsylvania State University, University Park, PA, United States

OPEN ACCESS

Edited by:

Jochen Steppan,
Johns Hopkins University,
United States

Reviewed by:

Eduardo Damasceno Costa,
Federal University of Minas Gerais,
Brazil

Yu Huang,
China University of Geosciences
Wuhan, China

Deepesh Pandey,
Johns Hopkins University,
United States

*Correspondence:

Sushant M. Ranadive
ranadive@umd.edu

Specialty section:

This article was submitted to
Vascular Physiology,
a section of the journal
Frontiers in Physiology

Received: 23 July 2021

Accepted: 25 August 2021

Published: 01 October 2021

Citation:

Ranadive SM, Dillon GA,
Mascone SE and Alexander LM
(2021) Vascular Health Triad
in Humans With Hypertension—Not
the Usual Suspects.
Front. Physiol. 12:746278.
doi: 10.3389/fphys.2021.746278

Hypertension (HTN) affects more than one-third of the US population and remains the top risk factor for the development of cardiovascular disease (CVD). Identifying the underlying mechanisms for developing HTN are of critical importance because the risk of developing CVD doubles with ~20 mmHg increase in systolic blood pressure (BP). Endothelial dysfunction, especially in the resistance arteries, is the primary site for initiation of sub-clinical HTN. Furthermore, inflammation and reactive oxygen and nitrogen species (ROS/RNS) not only influence the endothelium independently, but also have a synergistic influence on each other. Together, the interplay between inflammation, ROS and vascular dysfunction is referred to as the vascular health triad, and affects BP regulation in humans. While the interplay of the vascular health triad is well established, new underlying mechanistic targets are under investigation, including: Inducible nitric oxide synthase, hydrogen peroxide, hydrogen sulfide, nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB) and nuclear factor activated T cells. This review outlines the role of these *unusual suspects* in vascular health and function in humans. This review connects the dots using these *unusual suspects* underlying inflammation, ROS and vascular dysfunction especially in individuals at risk of or with diagnosed HTN based on novel studies performed in humans.

Keywords: blood pressure, endothelium, inflammation, oxidative stress, reactive oxygen species

INTRODUCTION

Cardiovascular disease (CVD) is the leading cause of morbidity and mortality worldwide, and the prevalence of CVD increases with aging (Virani et al., 2021). The increase in blood pressure (BP) or diagnosed hypertension (HTN) is widely accepted as the primary precursor to CVD; the risk of CVD is assumed to increase in a linear fashion as BP increases. In general, the risk of CVD doubles when there is an approximately 20 mmHg increase in systolic BP and 10 mmHg increase

in diastolic BP (Lewington et al., 2002). Further, BP rises substantially as humans age (Guzik and Touyz, 2017); however, the rise in BP with age is linear among men but rises in a differential pattern among women. Sex differences in HTN and their underlying mechanisms have been reviewed in detail previously (Ji et al., 2020).

In 2017, the American College of Cardiology (ACC) and American Heart Association (AHA) redefined the classifications of HTN diagnosis (Whelton et al., 2018). In the revised categorizations, the BP threshold has been lowered for stage 1 HTN and prehypertension (now elevated BP). Stage 1 HTN is defined as resting systolic BP between 130 and 139 mmHg or diastolic BP between 80 and 89 mmHg, and elevated BP is now defined as systolic BP between 120 and 129 mmHg and diastolic BP below 80 mmHg. Elevated BP is a strong predictor of late life HTN and CVD (Whelton et al., 2018). The revised categorizations have drastically increased the prevalence of HTN to about 46% of Americans (Virani et al., 2021), and have highlighted the importance of preclinical and clinical research in humans to identify therapeutic targets for interventions that extend the health of aging humans (Whelton et al., 2018). Moreover, identification of novel drug treatment based targets is important as medication adherence with traditional antihypertensives (diuretics, angiotensin converting enzyme inhibitors, calcium channel blockers, etc.) remains a significant issue.

The main site for vascular resistance, and thus a critical component of BP regulation, are the arterioles in the microcirculation (Nowroozpoor et al., 2021). The arterioles have similar anatomical layers as larger arteries; however, the lumen size of the arterioles (10–150 μm diameter) creates a substantial resistance to the blood flow and thus BP responses. In addition to the lumen size, the smooth muscle tone being normally in a state of contraction makes the resistance vessels an important site for BP control. Vascular resistance within the arteries is controlled by a complex interplay between local vasodilators, sympathetic modulation, and endocrine (paracrine) driven changes. Not only is the microvascular bed the first site to present with dysfunction, but it can also experience dysfunction without displaying any evidence in the macrovasculature or feed arteries (Nowroozpoor et al., 2021). Well established causes of endothelial dysfunction include imbalances in inflammation and/or reactive oxygen species (ROS). This is of particular importance because endothelial dysfunction can further potentiate imbalances in inflammation and ROS, leading to a “never ending cycle (or triad)” (Figure 1). The cumulative effects of these minor insults on the vasculature lead to a pro-hypertensive environment. In this mini-review we will first present aspects of the vascular health triad and then novel mechanisms that induce imbalances within the system, leading to HTN in humans. Table 1 summarizes the methods utilized to elucidate mechanisms underlying hypertension-associated vascular dysfunction. A comprehensive discussion of the renal and sympathetic contributions to HTN is beyond the scope of this review, but have been recently reviewed (Grassi et al., 2018, 2019; Fonkoue et al., 2019; Holwerda et al., 2019; Keir et al., 2020).

VASCULAR HEALTH—ROLE OF INFLAMMATION AND REACTIVE OXYGEN AND NITROGEN SPECIES—“THE USUAL SUSPECTS”

In young, healthy adults, inflammation and reactive oxygen and nitrogen species (ROS/RNS) serve a critical, positive physiological role in vascular homeostasis. The maintenance of vascular health is the complex relationship between vasoprotective factors, such as the nitric oxide system, and other pathways that impair these mechanisms, including both inflammation and ROS. “Normal” vascular function is often characterized by the ability to efficiently vasodilate or vasoconstrict in response to a stimulus. On the contrary, vascular dysfunction is characterized by the loss of efficiency in the vasodilatory component even in the presence of a stimulus. *Inflammation* is one such stimulus (and a cornerstone of the vascular health triad) which influences vascular function acutely and chronically—in a temporal fashion. Inflammation is heightened as a natural defense mechanism to tissue injury, infection, or pathogen infiltration. The initial inflammatory cascade is characterized by heightened pro-inflammatory cytokine release, immune cell movement to the site of invasion or injury, and the release of local chemoattractants, notably, adhesion molecules (Wadley et al., 2013). This acute inflammation (influx of inflammatory cytokines) can transiently impair vascular function (8–32 h), through an acute impairment in nitric oxide (NO) bioavailability, even in young otherwise healthy individuals (Hingorani et al., 2000). The second component of the vascular health triad is ROS, which are free radicals, such as superoxide or peroxynitrite, that are integral to cellular signaling (El Assar et al., 2013; Wadley et al., 2013). ROS are produced via oxidative metabolism and proteins, such as NADPH oxidase, xanthine oxidase and via endothelial nitric oxide synthase (eNOS) uncoupling (Jacobi et al., 2005; Ding et al., 2007; El Assar et al., 2013; Wadley et al., 2013). Endogenous antioxidants, such as superoxide dismutase (SOD), glutathione, and NO, clear ROS enzymatically, or through direct chemical reaction (Selemidis et al., 2007; El Assar et al., 2013; Wadley et al., 2013). The aspect of mitochondrial ROS is covered in detail in an excellent review by Kirkman et al. (2021).

In young, otherwise healthy adults, there is (*I*) redundancy in the vasodilatory pathways with abundant bioavailability of endothelium-derived vasodilating substances (e.g., NO, prostaglandins, EDHF) (*II*) low basal concentrations of vasoconstrictive substances and (*III*) low basal concentrations of inflammatory cytokines and ROS. However, while there is ample data indicating impaired endothelial function in human subject cohorts who are at risk for the development of HTN later in life, including those with a family history of HTN (Greaney et al., 2015; Matthews et al., 2017), and non-traditional CVD risk factors (Martens et al., 2016; Greaney et al., 2019; Katulka et al., 2019), there is lack of data in individuals who are young and otherwise healthy but have undiagnosed HTN. Moreover, due to this redundancy it is difficult to unravel these interrelated

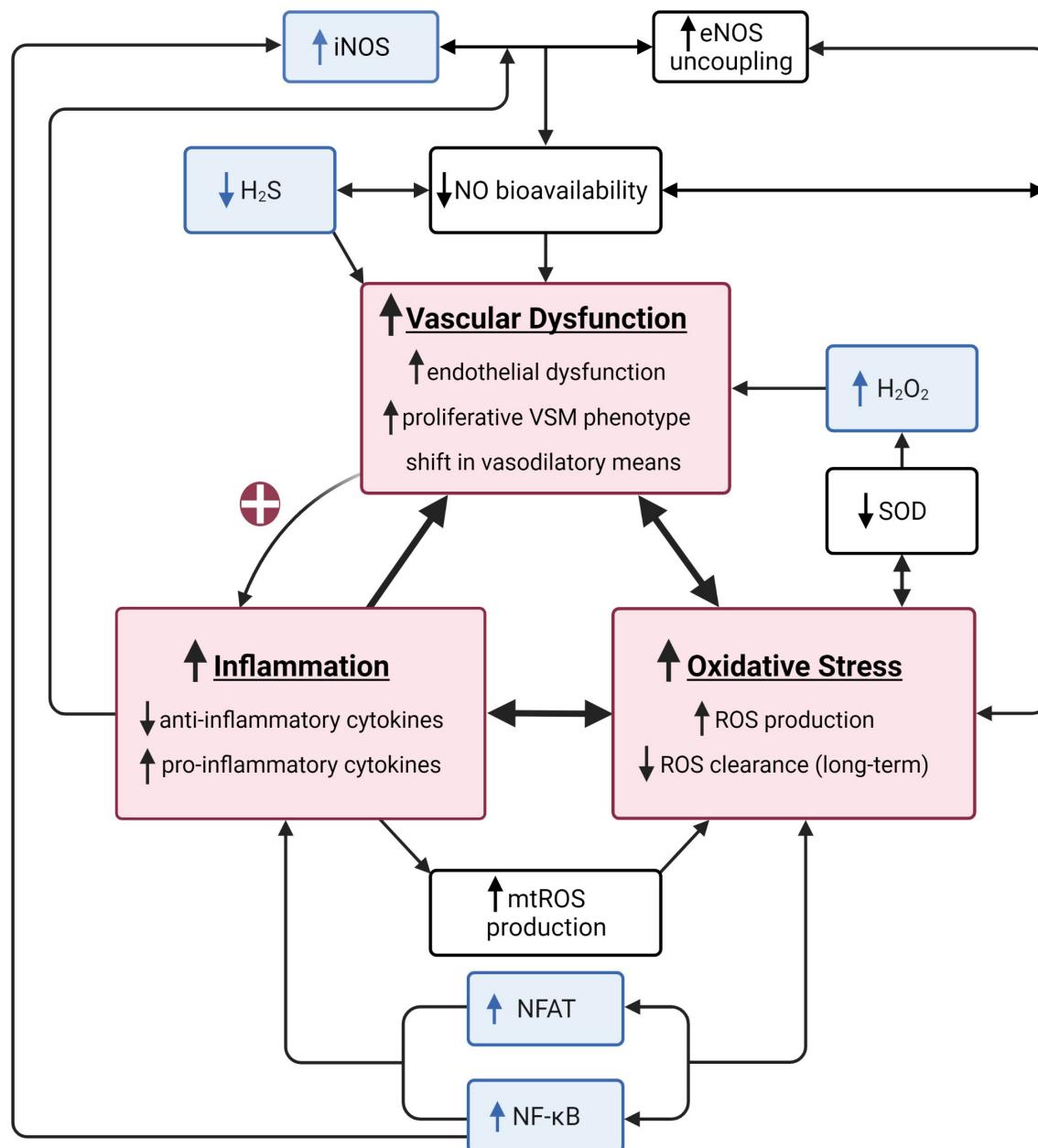


FIGURE 1 | A summary of the vascular health triad. The vascular health triad is composed of oxidative stress, inflammation, and vascular dysfunction (red boxes). These outcomes are synergistically interdependent as their underlying mechanisms directly (e.g., high inflammatory state causes a decrease in nitric oxide (NO) bioavailability resulting in increased vascular dysfunction) or indirectly [e.g., vascular dysfunction increasing inflammation via positive feedback loop (red plus sign)] interact, resulting in a vicious cycle of increased cardiovascular disease risk. Well-established underlying mechanisms of the triad include decreased NO bioavailability via increased endothelial nitric oxide synthase (eNOS) uncoupling, decreased superoxide dismutase (SOD), and increased mitochondrial reactive oxygen species (mtROS) production (white boxes). The unusual suspects include inducible nitric oxide synthase (iNOS), hydrogen sulfide (H₂S), hydrogen peroxide (H₂O₂), nuclear factor kappa-light-chain-enhancer of active B cells (NF-κB), and nuclear factor of activated T cells (NFAT; blue boxes). Created with BioRender.com.

mechanisms using gross measures of endothelial function in conduit arteries.

Our laboratory recently induced acute inflammation using an influenza vaccine *in vivo* and *in vitro* in young, healthy African American and Caucasian American individuals (Sapp et al., 2021). Although the vaccine stimulus did not impact conduit

artery function (as measured by flow-mediated dilation), there were decreases in eNOS messenger RNA in the African American group, coinciding with race-specific changes in intracellular and extracellular microRNAs (miR) related to inflammation (miR-221-3p, 222-3p, and 150-5p) (Sapp et al., 2021). Circulating miRs are novel biomarkers of acute and chronic inflammation

TABLE 1 | Summary of methodology for mechanisms mediating vascular function in hypertensive adults.

Method	References	Pathway
In vivo		
Flow-mediated Dilation + Doppler Ultrasound		
Gokce et al., 2001; Benjamin et al., 2004; Juonala et al., 2004; McGowan et al., 2006; Buus et al., 2007; Yang et al., 2010; Broxterman et al., 2019; Figueiredo et al., 2012	Endothelium-dependent dilation	
Ratchford et al., 2019a	Endothelium-dependent dilation Oxidative stress (acute antioxidant cocktail)	
Sapp et al., 2021	Endothelium-dependent dilation Inflammation (influenza vaccine)	
Craighead et al., 2020	Endothelium-dependent dilation, Endothelium in-dependent dilation (sublingual NTG), Oxidative stress (Vitamin C)	
Venous Occlusion Plethysmography		
On et al., 2002	Endothelium-dependent (ACh)	
Bottino et al., 2015	Endothelium-dependent dilation, Endothelium in-dependent (sublingual NTG)	
Hingorani et al., 2000	Endothelium-dependent (ACh, Bradykinin) Endothelium in-dependent (NTG, verapamil) Inflammation (salmonella typhi vaccine)	
Arterial Infusions + Doppler Ultrasound		
Takase et al., 2006; Broxterman et al., 2019	Endothelium-dependent dilation (ACh)	
Gokce et al., 2001; McGowan et al., 2006; Buus et al., 2007	Endothelium in-dependent dilation (NTG)	
Iontophoresis + Laser Doppler Flowmetry		
Cupisti et al., 2000	Endothelium-dependent dilation (ACh) Endothelium-independent dilation (SNP)	

TABLE 1 | (Continued)

Method	References	Pathway
In vivo		
Intradermal Microdialysis + Laser Doppler Flowmetry		
Farkas et al., 2004	Endothelium-dependent dilation (ACh; local heating)	
Lindstedt et al., 2006	Endothelium in-dependent dilation (SNP)	
Smith et al., 2011	Endothelium-dependent dilation (ACh, local heating) NO (L-NAME) iNOS (1400W) nNOS (NPLA)	
Smith et al., 2013	Vasoconstriction (local cooling, yohimbine + propranolol), Rho/Rho-Kinase (fasudil)	
Bruning et al., 2015	Endothelium-dependent dilation (local heating), NO (L-NAME)	
Craighead et al., 2017	Endothelium-dependent dilation (ACh, local heating), NO (L-NAME), iNOS (1400w)	
HTN Greaney et al., 2017	Endothelium-dependent dilation (ACh), NO (L-NAME), H ₂ S (Na2S, AOAA)	
Craighead et al., 2018	Endothelium independent dilation (SNP), NE-induced vasoconstriction, Lysyl Oxidation (BAPN)	
Dillon et al., 2020	Endothelium-dependent (ACh), NO (L-NAME)	
Sublingual NTG + Doppler Ultrasound		
McGowan et al., 2006	Endothelium in-dependent dilation	
Non-invasive Single Limb Exercise + Doppler Ultrasound		
Ratchford et al., 2019b	BF response to exercise (IHG)	
McGowan et al., 2006	BF response to exercise (static IHG, dynamic knee extension)	
Single Limb Exercise + NIRS-derived TSI		
Sprick et al., 2019	BF response to exercise (IHG)	

(Continued)

TABLE 1 |(Continued)

Method	References	Pathway
In vivo		
Ex vivo / in vitro Circulating concentrations		
	Hingorani et al., 2000	IL-1 β , IL-1Ra IL-6, TNF- α
	Bottino et al., 2015	IL-1, IL-6, OxLDL, sICAM, sVCAM, sE-Selectin, TNF- α
	Junqueira et al., 2018	Adiponectin, CRP, endothelin, ICAM, VCAM
	Craighead et al., 2020	IL-6, OxLDL, TNF- α , total antioxidant status, hsCRP
PBMCs		
	Huang et al., 2016	IL-6, NFAT, TNF- α
Arterial Biopsy		
	Phillips et al., 2007	NO (L-NAME) Oxidative stress (H ₂ O ₂)
	Donato et al., 2007	Oxidative stress (nitrotyrosine, NFkB)
	Migrino et al., 2011	Endothelial-dependent dilation (ACh), Endothelial in-dependent dilation (papaverine), Oxidative stress (SOD, BH4, mitoquinone, gp91ds-tat)
	Beyer et al., 2014	Oxidative stress (H ₂ O ₂ , mtROS, tempol)
	Beyer et al., 2017	Oxidative stress (H ₂ O ₂ , mtROS)
	Kadlec et al., 2017	H ₂ O ₂ , NO, PGC-1 α
	Hughes et al., 2021b	NO (L-NAME, c-PTIO), H ₂ O ₂ (peg-cat) Oxidative stress (rotenone)
	Hader et al., 2019	
	Hughes et al., 2021a	H ₂ O ₂
Venous Endothelial Cell Biopsy		
	Donato et al., 2007	Oxidative stress (NADPH oxidase p47phox, SOD, NFkB)
	Pierce et al., 2009	Inflammation (NFkB, TNF- α) Oxidative stress (NADPH oxidase p47phox)

(Continued)

TABLE 1 |(Continued)

Method	References	Pathway
In vivo		
	Craighead et al., 2020	Oxidative stress (NADPH, MnSOD), Inflammation (NFkB)
Cutaneous Biopsies		
	Smith et al., 2011	eNOS, iNOS, nNOS, pVASP
	Smith et al., 2013	ROCK activity and expression
	HTN Greaney et al., 2017	H ₂ S (CSE, 3-MPST)
Myograph		
	Buus et al., 2007	"small artery relaxation with ACh"

3-MPST, 3-mercaptopyruvate sulfurtransferase; ACh, acetylcholine; BAPN, β -aminopropionitrile; CRP, C-reactive protein; CSE, cystathione gamma-lyase; H₂S, hydrogen sulfide; hsCRP, high-sensitivity C-reactive protein; IHG, intermittent handgrip; IL, interleukin; iNOS, inducible nitric oxide synthase; L-NAME, NG-nitro-L-arginine methyl ester; MnSOD, manganese superoxide dismutase; NADPH, nicotinamide adenine dinucleotide phosphate; NE, norepinephrine; NFkB, nuclear factor kappa-light-chain-enhancer of activated B cells; nNOS, neuronal nitric oxide synthase; NO, nitric oxide; NPLA, N(ω)-propyl-L-arginine; NTG, nitroglycerin; oxLDL, oxidized low-density lipoprotein; PBMCs, peripheral blood mononuclear cells; ROCK, rho-associated protein kinase; ROS, reactive oxygen species; sICAM, soluble inter-cellular adhesion molecule; SNP, sodium nitroprusside; sVCAM, soluble vascular cell adhesion molecule; TNF- α , tumor necrosis factor alpha.

(Mi et al., 2013; Benz et al., 2016). miRs play mechanistic roles in endothelial activation, inflammation, and dysfunction; thus, initiating events in HTN (Cheng et al., 2014; Shi et al., 2015; Fernández-Hernando and Suárez, 2018). Furthermore, certain miRs [miR-146-protective (Wang et al., 2017); miR-34a- pro-inflammatory (Badi et al., 2018); miR-570-3p- pro-senescence (Baker et al., 2019); miR-217- pro-inflammatory (Zhang et al., 2019)] regulate ROS mechanisms via sirtuin-1 (Yamakuchi, 2012; Yamakuchi and Hashiguchi, 2018). Circulating miRs are taken up by endothelial cells, where they affect endothelial cell function, promote inflammation, and increase ROS (Njock and Fish, 2017; Zhong et al., 2018). Furthermore, the flow patterns within the arterial tree, i.e., turbulent flow near bifurcations or atherosclerotic lesions which can accentuate endothelial permeability or in contrast laminar flow which can exert beneficial effects on the vessel wall tend to trigger a number of miRs (Marin et al., 2013; Schmitz et al., 2019). Case in point, laminar shear stress (could be exercise induced) can upregulate miR-126 which is generally accepted as an anti-inflammatory and anti-atherogenic miR (Sapp et al., 2017). Therefore, in recent years circulating miRs have emerged as novel molecules mediating cell-to-cell communication in physiological processes (Njock and Fish, 2017; Bär et al., 2019). Mechanistically, emerging data in humans indicates significant cross-talk between inflammation and ROS within the endothelial cells in young, otherwise healthy adults, even before measurable conduit artery or macrovascular dysfunction plausibly through miRs (Sapp et al., 2021).

VASCULAR HEALTH TRIAD—AGING OR DISEASED STATES

In aging and diseased states, the vascular health triad becomes a positive feedback loop of heightened inflammation, oxidative stress, and vascular dysfunction. Chronic overproduction of inflammatory mediators, such as inflammatory cytokines, adhesion molecules, and inflammatory proteins, result in systemic low-grade inflammation initiating the complex cascade of heightened NADPH oxidase activity, eNOS uncoupling, and mitochondrial dysfunction, all of which result in heightened ROS production. Early in aging or disease pathogenesis, heightened antioxidant concentrations accommodate excessive ROS production. Following long-term exposure to increased ROS and inflammation, antioxidant defense mechanisms are reduced. Previous studies have interrogated these mechanisms with the acute administration of antioxidant cocktails (including ascorbic acid and alpha-tocopherol) in older adults (Eskurza et al., 2004a,b; Crecelius et al., 2010; Wray et al., 2012; Richards et al., 2015; Trinity et al., 2016), post-menopausal females (Ozemek et al., 2016), and individuals with heart failure with preserved ejection fraction (Ratchford et al., 2019a). However, more recent studies have focused on targeted pharmacological agents to identify specific ROS molecules within the triad (Alexander et al., 2013; Hurr et al., 2018; Martens et al., 2018; Park et al., 2018; Rossman et al., 2018). One emerging link in this triad is through the inducible nitric oxide synthase (iNOS) pathway. iNOS activity is increased during inflammation and NO is produced in toxic concentrations to prevent cell death by clearing excessive ROS (Aktan, 2004). NO reacts with superoxide (O_2^-), a main type of ROS produced during heightened inflammation, to form peroxynitrite ($ONOO^-$). Concurrently, superoxide dismutase (SOD) reacts with superoxide (O_2^-) to form hydrogen peroxide (H_2O_2). Interestingly H_2O_2 becomes the main contributing vasodilatory substance when NO bioavailability is decreased (Phillips et al., 2007; Beyer et al., 2014; Kadlec et al., 2017). Initially with acute inflammatory/ROS stimuli, other vasodilatory substances, such as prostaglandins and H_2O_2 , can compensate for the impairment in NO-mediated vasodilation (Beyer et al., 2017). However, if the allostatic load presented by inflammation and ROS persists, not only do the compensatory mechanisms fail, but eventually deleterious vascular remodeling occurs. Therefore, with aging and/or disease progression the healthy functioning of the vasculature is disrupted due to (I) loss of redundant vasodilatory pathways, with limited bioavailability of vasodilating substances (especially NO), (II) increased circulating vasoconstrictive substances and (III) increased concentrations of inflammatory cytokines and ROS.

Typically, when considering vascular dysfunction, the role of NO bioavailability/scavenging due to eNOS uncoupling, decreased superoxide dismutase (SOD), and increased mitochondrial reactive oxygen species (mtROS) production are of significant importance (Figure 1). Therefore, in the following sections we have presented some of the newer and not widely discussed factors which can also have role in decreased NO bioavailability and/or increased NO scavenging, leading to vascular dysfunction.

VASCULAR HEALTH TRIAD—“THE UNUSUAL SUSPECTS”

Role of Inducible Nitric Oxide Synthase

During heightened inflammation and oxidative stress, there are various intersecting pathways that contribute to impaired NO bioavailability and, eventually, vascular dysfunction. Increased iNOS expression is stimulated by nuclear factor kappa-light-chain-enhancer of activated B cells (NF- κ B), interleukin-6 (IL-6), and ROS producers, such as the p47 phox subunit of NADPH oxidases (Hemmrich et al., 2003; Wu et al., 2008; Li et al., 2015). With chronic low-grade inflammation, iNOS activity increases, resulting in increased iNOS-derived NO production. To pharmacodissect the NOS balance in humans and prominent role of iNOS, we performed a unique *bed to bench* experiment evaluating endothelial-dependent microvascular function in individuals with HTN during iNOS, neuronal NOS (nNOS) and non-selective-NOS inhibition (Smith et al., 2011). Interestingly, the attenuated endothelial-dependent vasodilation in hypertensive adults was restored with iNOS inhibition, suggesting a prominent role of iNOS in hypertension-induced microvascular dysfunction. Even though the eNOS expression was similar between normotensives and hypertensives adults, iNOS expression in biopsy samples from hypertensive subjects was significantly greater as compared to age-matched normotensives (Smith et al., 2011). Thus, the NO produced during inflammation with HTN adopts a scavenging and cell-preserving role, as opposed to an active vasodilatory role.

Hydrogen Peroxide (H_2O_2) and H_2O_2 -Mediated Primary Vasodilatory Mechanism

SOD is a primary antioxidant defense system with three known forms: SOD1 (cytosolic), SOD2 (mitochondrial), and SOD3 (circulating) (Zelko et al., 2002). SOD scavenges O_2^- radicals to form H_2O_2 . During states of heightened inflammation and ROS, NADPH oxidase activity and eNOS uncoupling subsequently increase, resulting in increased O_2^- production (El Assar et al., 2013; Risbano and Gladwin, 2013; Wadley et al., 2013). In early disease initiation and progression, SOD activity increases to clear excessively produced O_2^- , subsequently increasing the production of H_2O_2 during both acute and chronic increases in inflammation and oxidative stress (El Assar et al., 2013). Recent studies from the Guterman laboratory have suggested a novel theory of shifts in vasodilatory pathways during vascular inflammatory conditions, especially HTN (Migrino et al., 2011; Beyer et al., 2014, 2017; Kadlec et al., 2017). The NO-mediated vasodilatory pathway is vasoprotective, as it helps maintain normal BP in young, healthy individuals. However, with increased intraluminal pressure, as seen in HTN and reduced bioavailability of NO, there is a shift in microvascular vasodilatory pathways toward H_2O_2 -dependent mechanisms, even though the total magnitude of the vasodilation to a given shear stimulus remains the same (Beyer et al., 2014). Hughes et al. (2021b) examined the effect of transient increases in intraluminal

pressure in resistance arterioles of hypertensive individuals with and without coronary artery disease were evaluated. In this model, there was a compensatory switch to the H₂O₂-mediated vasodilatory pathway following increased intraluminal pressure, suggesting diseased and healthy aged adults have similar shifts in primary vasodilatory mechanisms, but along a different time course (Hughes et al., 2021b). Furthermore, in the presence of transient increases in intraluminal pressure, even in isolated arterioles of healthy individuals, H₂O₂ is typically mitochondria derived (Beyer et al., 2014). These *ex-vivo* studies have also led to potential targets, such as autophagy and extranuclear telomerase (Hughes et al., 2021a). Taken together, these studies suggests an important switch in the physiological mechanism of vasodilation and heightened ROS production, specifically production of O₂⁻, H₂O₂, and ONOO⁻ as propagating a positive feedback loop that promotes vascular dysfunction.

Nuclear Factor Kappa-Light-Chain-Enhancer of Activated B Cells and Nuclear Factor From Activated T Cells

Investigating upstream transcriptional molecular targets underlying endothelial and vascular smooth muscle cell dysfunction in humans has yielded significant understanding of the complexity of these signaling mechanisms. Despite the complexity and multiple downstream effects, these studies are necessary for the development of targeted treatment and prevention strategies for HTN in humans. It is well established that NF-κB are an important intracellular mediator of inflammation and vascular dysfunction (specifically, NO-mediated mechanisms) (Donato et al., 2007, 2009; Pierce et al., 2009; Lee et al., 2014). In an elegant series of preclinical and clinical studies, Donato et al. (2009) established that activation of NF-κB mediates age-related vascular dysfunction. These studies interrogated NF-κB functionality in human subjects using a short-term (4 days) high-dose oral salsalate approach (Pierce et al., 2009). Salsalate, a non-acetylated salicylate, inhibits NF-κB translocation to the nucleus, reducing ROS synthesis through NADPH oxidases (Kopp and Ghosh, 1994; Pierce et al., 1996). They demonstrated that oral salsalate reduces endothelial cell NF-κB p65 expression by ~25%, total nitrotyrosine, a global marker of oxidative stress, and NADPH oxidase p47^{phox} expression by 25 and 30%, respectively. Importantly, inhibition of NF-κB functionally resulted in improved NO-dependent vasodilation. Additionally, when comparing inactive to habitually active older adults, active adults had reduced NF-κB p65 expression, reduced nitrotyrosine, and endothelial function similar to their younger counterparts (Walker et al., 2014).

A growing body of literature suggests that the family of Ca²⁺/calcineurin-sensitive transcriptional factors of nuclear factor from activated T-cells (NFAT) may play an essential role as a molecular switch that initiates dysfunction in both the endothelium (Cockerill et al., 1995; Boss et al., 1998; Armesilla

et al., 1999; Bochkov et al., 2002; Gonzalez Bosc et al., 2004; Zetterqvist et al., 2015; Huang et al., 2016) and vascular smooth muscle (Suzuki et al., 2002; Lipskaia et al., 2003; Amberg et al., 2004; Nilsson et al., 2006; Nieves-Cintrón et al., 2007; Orr et al., 2009; Pang and Sun, 2009; Nilsson-Berglund et al., 2010; Berglund et al., 2012; Shiny et al., 2016; Soudani et al., 2016; Govatati et al., 2019). NFATs regulate multiple downstream mechanisms that initiate vascular dysfunction. Specifically, NFATs (**I**) impair endothelial function through NO-dependent mechanisms (Armesilla et al., 1999; Bochkov et al., 2002; Johnson et al., 2003; Norata et al., 2007; Garcia-Vaz et al., 2020; Wang et al., 2020), (**II**) increase the expression of inflammatory mediators in the arterial wall promoting atherosclerosis (Pierce et al., 1996; Pang and Sun, 2009; Nilsson-Berglund et al., 2010; Berglund et al., 2012; Zetterqvist et al., 2014; Weng et al., 2017), and (**III**) initiate pathogenic VSM proliferation (Suzuki et al., 2002; Lipskaia et al., 2003; Nilsson et al., 2006; Nieves-Cintrón et al., 2007; Donato et al., 2009; Orr et al., 2009; Pang and Sun, 2009; Nilsson-Berglund et al., 2010; Yan et al., 2015; Shiny et al., 2016; Soudani et al., 2016; Govatati et al., 2019). In preclinical models, inhibition of NFAT has prevented the activation of inflammatory cytokines (Kiani et al., 2000; Zetterqvist et al., 2014; Bretz et al., 2015; Huang et al., 2016), enhanced eNOS expression (Smith et al., 2011), increased NO bioavailability (Friedman et al., 2014; Zetterqvist et al., 2014, 2015; Garcia-Vaz et al., 2020), prevented VSM proliferation (Lipskaia et al., 2003; Berglund et al., 2012; Shiny et al., 2016), lowered BP (Garcia-Vaz et al., 2020), and reduced total atherosclerotic load (Norata et al., 2007; Nilsson-Berglund et al., 2010; Zetterqvist et al., 2014). Currently, these investigations/findings are limited to cellular and animal models. There are promising approaches, including examining NFATs in the skin microcirculation and in peripheral blood mononuclear cells, for investigating the role of NFATs in humans (Huang et al., 2016). Elucidation of the role of NFATs and their putative upstream contributions to the vascular health triad in humans is still needed.

Hydrogen Sulfide

As one of the three gasotransmitters ubiquitously synthesized in mammalian systems, hydrogen sulfide (H₂S) is emerging as a critical component of vascular homeostasis (Polhemus and Lefer, 2014). HTN-associated microvascular dysfunction is characterized by a loss of endothelium-dependent signaling pathways, including hydrogen sulfide (H₂S) (Cupisti et al., 2000; Lindstedt et al., 2006; Smith et al., 2011; Craighead et al., 2017; Greaney et al., 2017). Similar to NO, H₂S exerts several beneficial physiological effects, including inhibiting inflammatory markers and leukocyte adhesion molecules, enhancing anti-inflammatory markers, and acting as an antioxidant (Polhemus and Lefer, 2014). NO and H₂S vasodilatory pathways are synergistically interdependent. Both exogenous and endogenous enzymatic H₂S synthesis helps to maintain NO bioavailability by stabilizing the eNOS dimer and improving tetrahydrobiopterin (BH₄) bioavailability (Zhao et al., 2001; Coletta et al., 2012;

Altaany et al., 2013; Polhemus and Lefer, 2014; Greaney et al., 2017). Exogenous administration of H₂S improves NO-dependent vasodilation, while blockade of the H₂S producing enzyme, cystathionine γ-layase (CSE), impairs NO-dependent vasodilation (Coletta et al., 2012; Altaany et al., 2013). Reciprocally, blockade of NO synthesis also reduces H₂S-dependent vasodilation (Zhao et al., 2001; Coletta et al., 2012). We reported that the H₂S-dependent contribution to endothelium-dependent vasodilation is functionally absent in naïve-to-therapy hypertensive adults (Greaney et al., 2017). This was partially due to reduced endogenous enzymatic synthesis of H₂S, as expression and activity of H₂S producing enzymes, including cystathione-γ-lyase and 3-mercaptopryruvate transulferase, were reduced in hypertensive compared to normotensive adults (Greaney et al., 2017). However, vascular responsiveness to exogenous H₂S donors remained intact in hypertensive adults. In preclinical models, treatment with an H₂S donating antihypertensive agents improves endothelial function and normalizes BP (Bucci et al., 2014; Ji et al., 2014; Al-Magableh et al., 2015; Xue et al., 2015). These improvements were partially mediated by increased NO bioavailability (Bucci et al., 2014; Ji et al., 2014; Al-Magableh et al., 2015; Xue et al., 2015), demonstrating the synergistic nature of these gasotransmitter pathways. Various nutraceutical intervention studies suggest that allicin, the bioactive component of garlic, improves vascular function, specifically through the H₂S enzymatic pathway (Cui et al., 2020). Due to the ubiquitous nature of H₂S, there are several clinical trials evaluating the impact of H₂S donating pharmacologics in a variety of disease states. At present, the H₂S synthetic pathway remains an underexplored therapeutic target in human HTN and other CVDs, including heart failure.

Potential Targets

In humans, the potential targets to mitigate the vascular health triad and improve the vascular function have been pursued from a global, holistic approach of exercise training to more specific, targeted treatments. In humans, ascorbic acid and folic acid supplementation have been shown to improve vascular function in populations with inflammatory diseases (Alexander et al., 2013; Karbach et al., 2014; Stanhewicz et al., 2015; Stanhewicz and Kenney, 2017). BH₄ precursors and antioxidants are the main therapeutic targets facilitating eNOS coupling and reducing eNOS-derived ROS production. BH₄ in the form of saproterin is an orphan drug used in the treatment of certain genetic variants of phenylketonuria. Saproterin has improved eNOS function and NO-dependent vasodilation in aged and hypercholesterolemic human subjects in both acute and interventional studies. Specifically, saproterin (or, BH₄) supplementation works through eNOS coupling mechanisms and

not simply through its moderate antioxidant capacity. Folic acid (and its active metabolite 5-MTHF) is a cost-effective strategy for improving BH₄ bioavailability through BH₂ recycling. Thus, reducing eNOS uncoupling is an attractive, accessible, and affordable intervention for improving the vascular health triad in aging and diseased states.

Similarly, mitochondrial ROS targeted interventions have shown promise in the past decade. Nicotinamide riboside (NR) is a sirtuin-1 (SIRT-1) precursor that has gained recent popularity in vascular intervention treatment research. NR is a precursor to NAD⁺, and SIRT1 is a NAD⁺-dependent deacetylase. Similar to resveratrol and Mito Q, NR supplementation has had vastly positive impacts on vascular function and oxidative stress in pre-clinical animal models (Yoshino et al., 2011; Mills et al., 2016). NR has been shown to improve vascular function and reduce oxidative stress in aged mice. However, in humans, these findings have not been replicated. NR has shown small impacts on various measures of vascular function (Martens et al., 2018). Thus, there is a knowledge gap in the positive benefits of long-term NR supplementation.

CONCLUSION

In summary, there is ample evidence that the shift from healthy endothelial function to dysfunction, typically preceding HTN and CVD, is driven by a cross-talk between inflammation and ROS. There are numerous “players” that have been recently identified to be responsible for this abnormal shift toward dysfunction. Therefore, it is crucial to investigate these key players in human experimental models to further understand and improve human vascular function with the ultimate goal of preventing CVD risk.

AUTHOR CONTRIBUTIONS

SR and LA conceived the research review. SR, GD, SM, and LA designed the research review, drafted, revised, and approved the final version of the manuscript. GD and SM prepared figure and table. All authors contributed to the article and approved the submitted version.

FUNDING

Support for this work was provided by National Institute of Health (NIH) grant T32-5T32-AG049676 to GD, NIH grant R01-HL-093238 to LA, and University of Maryland (UMD) Tier 1 (pilot grant) to SR.

REFERENCES

- Aktan, F. (2004). iNOS-mediated nitric oxide production and its regulation. *Life Sci.* 75, 639–653. doi: 10.1016/j.lfs.2003.10.042
 Alexander, L. M., Kutz, J. L., and Kenney, W. L. (2013). Tetrahydrobiopterin increases NO-dependent vasodilation in hypercholesterolemic human skin

- through eNOS-coupling mechanisms. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 304, R164–R169. doi: 10.1152/ajpregu.00448.2012
 Al-Magableh, M. R., Kemp-Harper, B. K., and Hart, J. L. (2015). Hydrogen sulfide treatment reduces blood pressure and oxidative stress in angiotensin II-induced hypertensive mice. *Hypertens. Res.* 38, 13–20. doi: 10.1038/hr.014.125

- Altaany, Z., Yang, G., and Wang, R. (2013). Crosstalk between hydrogen sulfide and nitric oxide in endothelial cells. *J. Cell. Mol. Med.* 17, 879–888. doi: 10.1111/jcmm.12077
- Amberg, G. C., Rossow, C. F., Navedo, M. F., and Santana, L. F. (2004). NFATc3 regulates Kv2.1 expression in arterial smooth muscle. *J. Biol. Chem.* 279, 47326–47334. doi: 10.1074/jbc.M408789200
- Armesilla, A. L., Lorenzo, E., Gomez del Arco, P., Martinez-Martinez, S., Alfranca, A., and Redondo, J. M. (1999). Vascular endothelial growth factor activates nuclear factor of activated T cells in human endothelial cells: a role for tissue factor gene expression. *Mol. Cell Biol.* 19, 2032–2043. doi: 10.1128/MCB.19.3.2032
- Badi, I., Mancinelli, L., Polizzotto, A., Ferri, D., Zeni, F., Burba, I., et al. (2018). miR-34a Promotes Vascular Smooth Muscle Cell Calcification by Downregulating SIRT1 (Sirtuin 1) and Axl (AXL Receptor Tyrosine Kinase). *Arterioscler. Thromb. Vascul. Biol.* 38, 2079–2090. doi: 10.1161/ATVBAHA.118.311298
- Baker, J. R., Vuppusetty, C., Colley, T., Hassibi, S., Fenwick, P. S., Donnelly, L. E., et al. (2019). MicroRNA-570 is a novel regulator of cellular senescence and inflammasome. *FASEB J.* 33, 1605–1616. doi: 10.1096/fj.20180965R
- Bär, C., Thum, T., and de Gonzalo-Calvo, D. (2019). Circulating miRNAs as mediators in cell-to-cell communication. *Epigenomics* 11, 111–113. doi: 10.2217/epi-2018-0183
- Benjamin, E. J., Larson, M. G., Keyes, M. J., Mitchell, G. F., Vasan, R. S., Keaney, J. F. Jr., et al. (2004). Clinical correlates and heritability of flow-mediated dilation in the community: the Framingham Heart Study. *Circulation* 109, 613–619. doi: 10.1161/01.CIR.0000112565.60887.1E
- Benz, F., Roy, S., Trautwein, C., Roderburg, C., and Luedde, T. (2016). Circulating MicroRNAs as Biomarkers for Sepsis. *Int. J. Mol. Sci.* 17:78. doi: 10.3390/ijms17010078
- Berglund, L. M., Kotova, O., Osmark, P., Grufman, H., Xing, C., Lydrup, M. L., et al. (2012). NFAT regulates the expression of AIF-1 and IRT-1: yin and yang splice variants of neointima formation and atherosclerosis. *Cardiovasc. Res.* 93, 414–423. doi: 10.1093/cvr/cvr309
- Beyer, A. M., Durand, M. J., Hockenberry, J., Gamblin, T. C., Phillips, S. A., and Guterman, D. D. (2014). An acute rise in intraluminal pressure shifts the mediator of flow-mediated dilation from nitric oxide to hydrogen peroxide in human arterioles. *Am. J. Physiol. Heart Circ. Physiol.* 307, H1587–H1593. doi: 10.1152/ajpheart.00557.2014
- Beyer, A. M., Zinkevich, N., Miller, B., Liu, Y., Wittenburg, A. L., Mitchell, M., et al. (2017). Transition in the mechanism of flow-mediated dilation with aging and development of coronary artery disease. *Basic Res. Cardiol.* 112:5. doi: 10.1007/s00395-016-0594-x
- Bochkov, V. N., Mechthaleriakova, D., Lucerna, M., Huber, J., Malli, R., Graier, W. F., et al. (2002). Oxidized phospholipids stimulate tissue factor expression in human endothelial cells via activation of ERK/EGR-1 and Ca(++)/NFAT. *Blood* 99, 199–206. doi: 10.1182/blood.V99.1.199
- Boss, V., Wang, X., Koppelman, L. F., Xu, K., and Murphy, T. J. (1998). Histamine induces nuclear factor of activated T cell-mediated transcription and cyclosporin A-sensitive interleukin-8 mRNA expression in human umbilical vein endothelial cells. *Mol. Pharmacol.* 54, 264–272. doi: 10.1124/mol.54.2.264
- Bottino, D. A., Lopes, F. G., de Oliveira, F. J., Mecenas, A. S., Clapauch, R., and Bouskela, E. (2015). Relationship between biomarkers of inflammation, oxidative stress and endothelial/microcirculatory function in successful aging versus healthy youth: a transversal study. *BMC Geriatr.* 15:41. doi: 10.1186/s12877-015-0044-x
- Bretz, C. A., Savage, S. R., Capozzi, M. E., Suarez, S., and Penn, J. S. (2015). NFAT isoforms play distinct roles in TNFalpha-induced retinal leukostasis. *Sci. Rep.* 5:14963. doi: 10.1038/srep14963
- Broxterman, R. M., Witman, M. A., Trinity, J. D., Groot, H. J., Rossman, M. J., Park, S. Y., et al. (2019). Strong Relationship Between Vascular Function in the Coronary and Brachial Arteries. *Hypertension* 74, 208–215. doi: 10.1161/HYPERTENSIONAHA.119.12881
- Bruning, R. S., Kenney, W. L., and Alexander, L. M. (2015). Altered skin flowmotion in hypertensive humans. *Microvascular. Res.* 97, 81–87. doi: 10.1016/j.mvr.2014.01.001
- Bucci, M., Vellecco, V., Cantalupo, A., Brancaleone, V., Zhou, Z., Evangelista, S., et al. (2014). Hydrogen sulfide accounts for the peripheral vascular effects of zofenopril independently of ACE inhibition. *Cardiovasc. Res.* 102, 138–147. doi: 10.1093/cvr/cvu026
- Buus, N. H., Jørgensen, C. G., Mulvany, M. J., and Sørensen, K. E. (2007). Large and small artery endothelial function in patients with essential hypertension—effect of ACE inhibition and beta-blockade. *Blood Press.* 16, 106–113. doi: 10.1080/08037050701343688
- Cheng, H. S., Njock, M. S., Khuzha, N., Dang, L. T., and Fish, J. E. (2014). Noncoding RNAs regulate NF-κB signaling to modulate blood vessel inflammation. *Front. Genet.* 5:422. doi: 10.3389/fgene.2014.00422
- Cockerill, G. W., Bert, A. G., Ryan, G. R., Gamble, J. R., Vadas, M. A., and Cockerill, P. N. (1995). Regulation of granulocyte-macrophage colony-stimulating factor and E-selectin expression in endothelial cells by cyclosporin A and the T-cell transcription factor NFAT. *Blood* 86, 2689–2698. doi: 10.1182/blood.V86.7.2689.2689
- Coletta, C., Papapetropoulos, A., Erdelyi, K., Olah, G., Modis, K., Panopoulos, P., et al. (2012). Hydrogen sulfide and nitric oxide are mutually dependent in the regulation of angiogenesis and endothelium-dependent vasorelaxation. *Proc. Natl. Acad. Sci. U S A.* 109, 9161–9166. doi: 10.1073/pnas.1202916109
- Craighead, D. H., Freeberg, K. A., and Seals, D. R. (2020). Vascular Endothelial Function in Midlife/Older Adults Classified According to 2017 American College of Cardiology/American Heart Association Blood Pressure Guidelines. *J. Am. Heart Assoc.* 9:e016625. doi: 10.1161/JAHA.120.016625
- Craighead, D. H., Smith, C. J., and Alexander, L. M. (2017). Blood pressure normalization via pharmacotherapy improves cutaneous microvascular function through NO-dependent and NO-independent mechanisms. *Microcirculation* 24:12382. doi: 10.1111/micc.12382
- Craighead, D. H., Wang, H., Santhanam, L., and Alexander, L. M. (2018). Acute lysyl oxidase inhibition alters microvascular function in normotensive but not hypertensive men and women. *Am. J. Physiol. Heart Circ. Physiol.* 314, H422–H433. doi: 10.1152/ajpheart.00521.2017
- Creelius, A. R., Kirby, B. S., Voyles, W. F., and Dinrenno, F. A. (2010). Nitric oxide, but not vasodilating prostaglandins, contributes to the improvement of exercise hyperemia via ascorbic acid in healthy older adults. *Am. J. Physiol. Heart Circ. Physiol.* 299, H1633–H1641. doi: 10.1152/ajpheart.00614.2010
- Cui, T., Liu, W., Chen, S., Yu, C., Li, Y., and Zhang, J. Y. (2020). Antihypertensive effects of allicin on spontaneously hypertensive rats via vasorelaxation and hydrogen sulfide mechanisms. *Biomed. Pharmacother.* 128:110240. doi: 10.1016/j.biopharm.2020.110240
- Cupisti, A., Rossi, M., Placidi, S., Fabbri, A., Morelli, E., Vagheggi, G., et al. (2000). Responses of the skin microcirculation to acetylcholine in patients with essential hypertension and in normotensive patients with chronic renal failure. *Nephron* 85, 114–119. doi: 10.1159/000045643
- Dillon, G. A., Greaney, J. L., Shank, S., Leuenberger, U. A., and Alexander, L. M. (2020). AHA/ACC-defined stage 1 hypertensive adults do not display cutaneous microvascular endothelial dysfunction. *Am. J. Physiol. Heart Circ. Physiol.* 319, H539–H546. doi: 10.1152/ajpheart.00179.2020
- Ding, H., Aljofan, M., and Triggle, C. R. (2007). Oxidative stress and increased eNOS and NADPH oxidase expression in mouse microvessel endothelial cells. *J. Cell Physiol.* 212, 682–689. doi: 10.1002/jcp.21063
- Donato, A. J., Eskurza, I., Silver, A. E., Levy, A. S., Pierce, G. L., Gates, P. E., et al. (2007). Direct evidence of endothelial oxidative stress with aging in humans: relation to impaired endothelium-dependent dilation and upregulation of nuclear factor-kappaB. *Circ. Res.* 100, 1659–1666. doi: 10.1161/01.RES.0000269183.13937.e8
- Donato, A. J., Pierce, G. L., Lesniewski, L. A., and Seals, D. R. (2009). Role of NFκB in age-related vascular endothelial dysfunction in humans. *Aging* 1, 678–680. doi: 10.18632/aging.100080
- El Assar, M., Angulo, J., and Rodriguez-Manas, L. (2013). Oxidative stress and vascular inflammation in aging. *Free Radic. Biol. Med.* 65, 380–401. doi: 10.1016/j.freeradbiomed.2013.07.003
- Eskurza, I., Monahan, K. D., Robinson, J. A., and Seals, D. R. (2004a). Ascorbic acid does not affect large elastic artery compliance or central blood pressure in

- young and older men. *Am. J. Physiol. Heart Circ. Physiol.* 286, H1528–H1534. doi: 10.1152/ajpheart.00879.2003
- Eskurza, I., Monahan, K. D., Robinson, J. A., and Seals, D. R. (2004b). Effect of acute and chronic ascorbic acid on flow-mediated dilatation with sedentary and physically active human ageing. *J. Physiol.* 556(Pt 1), 315–324. doi: 10.1113/jphysiol.2003.057042
- Farkas, K., Kolossvary, E., Järai, Z., Nemcsik, J., and Farsang, C. (2004). Non-invasive assessment of microvascular endothelial function by laser Doppler flowmetry in patients with essential hypertension. *Atherosclerosis* 173, 97–102. doi: 10.1016/j.atherosclerosis.2003.11.015
- Fernández-Hernando, C., and Suárez, Y. (2018). MicroRNAs in endothelial cell homeostasis and vascular disease. *Curr. Opin. Hematol.* 25, 227–236. doi: 10.1097/MOH.0000000000000424
- Figueiredo, V. N., Yugar-Toledo, J. C., Martins, L. C., Martins, L. B., de Faria, A. P., de Haro Moraes, C., et al. (2012). Vascular stiffness and endothelial dysfunction: Correlations at different levels of blood pressure. *Blood Press.* 21, 31–38. doi: 10.3109/08037051.2011.617045
- Fonkoue, I. T., Le, N. A., Kankam, M. L., DaCosta, D., Jones, T. N., Marvar, P. J., et al. (2019). Sympathoexcitation and impaired arterial baroreflex sensitivity are linked to vascular inflammation in individuals with elevated resting blood pressure. *Physiol. Rep.* 7:e14057. doi: 10.14814/phy2.14057
- Friedman, J. K., Nitta, C. H., Henderson, K. M., Codianni, S. J., Sanchez, L., Ramiro-Diaz, J. M., et al. (2014). Intermittent hypoxia-induced increases in reactive oxygen species activate NFATc3 increasing endothelin-1 vasoconstrictor reactivity. *Vascul. Pharmacol.* 60, 17–24. doi: 10.1016/j.vph.2013.11.001
- Garcia-Vaz, E., McNeilly, A. D., Berglund, L. M., Ahmad, A., Gallagher, J. R., Dutius Andersson, A. M., et al. (2020). Inhibition of NFAT Signaling Restores Microvascular Endothelial Function in Diabetic Mice. *Diabetes* 69, 424–435. doi: 10.2337/db18-0870
- Gokce, N., Holbrook, M., Duffy, S. J., Demissie, S., Cupples, L. A., Biegelsen, E., et al. (2001). Effects of race and hypertension on flow-mediated and nitroglycerin-mediated dilation of the brachial artery. *Hypertension* 38, 1349–1354. doi: 10.1161/hy1201.096575
- Gonzalez Bosc, L. V., Wilkerson, M. K., Bradley, K. N., Eckman, D. M., Hill-Eubanks, D. C., and Nelson, M. T. (2004). Intraluminal pressure is a stimulus for NFATc3 nuclear accumulation: role of calcium, endothelium-derived nitric oxide, and cGMP-dependent protein kinase. *J. Biol. Chem.* 279, 10702–10709. doi: 10.1074/jbc.M312920200
- Govatati, S., Pichavaram, P., Janjanam, J., Zhang, B., Singh, N. K., Mani, A. M., et al. (2019). NFATc1-E2F1-LMCD1-Mediated IL-33 Expression by Thrombin Is Required for Injury-Induced Neointima Formation. *Arterioscler. Thromb. Vascul. Biol.* 39, 1212–1226. doi: 10.1161/ATVBAHA.119.312729
- Grassi, G., Biffi, A., Seravalle, G., Trevano, F. Q., Dell’Oro, R., Corrao, G., et al. (2019). Sympathetic Neural Overdrive in the Obese and Overweight State. *Hypertension* 74, 349–358. doi: 10.1161/HYPERTENSIONAHA.119.12885
- Grassi, G., Pisano, A., Bolignano, D., Seravalle, G., D’Arrigo, G., Quartu-Trevano, F., et al. (2018). Sympathetic Nerve Traffic Activation in Essential Hypertension and Its Correlates: Systematic Reviews and Meta-Analyses. *Hypertension* 72, 483–491. doi: 10.1161/HYPERTENSIONAHA.118.11038
- Greaney, J. L., Kutz, J. L., Shank, S. W., Jandu, S., Santhanam, L., and Alexander, L. M. (2017). Impaired Hydrogen Sulfide-Mediated Vasodilation Contributes to Microvascular Endothelial Dysfunction in Hypertensive Adults. *Hypertension* 69, 902–909. doi: 10.1161/HYPERTENSIONAHA.116.08964
- Greaney, J. L., Matthews, E. L., and Wenner, M. M. (2015). Sympathetic reactivity in young women with a family history of hypertension. *Am. J. Physiol. Heart Circ. Physiol.* 308, H816–H822. doi: 10.1152/ajpheart.00867.2014
- Greaney, J. L., Saunders, E. F. H., Santhanam, L., and Alexander, L. M. (2019). Oxidative Stress Contributes to Microvascular Endothelial Dysfunction in Men and Women With Major Depressive Disorder. *Circ. Res.* 124, 564–574. doi: 10.1161/CIRCRESAHA.118.313764
- Guzik, T. J., and Touyz, R. M. (2017). Oxidative Stress, Inflammation, and Vascular Aging in Hypertension. *Hypertension* 70, 660–667. doi: 10.1161/HYPERTENSIONAHA.117.07802
- Hader, S. N., Zinkevich, N., Norwood Toro, L. E., Kriegel, A. J., Kong, A., Freed, J. K., et al. (2019). Detrimental effects of chemotherapy on human coronary microvascular function. *Am. J. Physiol. Heart Circ. Physiol.* 317, H705–H710. doi: 10.1152/ajpheart.00370.2019
- Hemmrich, K., Suschek, C. V., Lerzynski, G., and Kolb-Bachofen, V. (2003). iNOS activity is essential for endothelial stress gene expression protecting against oxidative damage. *J. Appl. Physiol.* 95, 1937–1946. doi: 10.1152/japplphysiol.00419.2003
- Hingorani, A. D., Cross, J., Kharbanda, R. K., Mullen, M. J., Bhagat, K., Taylor, M., et al. (2000). Acute systemic inflammation impairs endothelium-dependent dilatation in humans. *Circulation* 102, 994–999. doi: 10.1161/01.CIR.102.9.994
- Holwerda, S. W., Luehrs, R. E., DuBose, L., Collins, M. T., Wooldridge, N. A., Stroud, A. K., et al. (2019). Elevated Muscle Sympathetic Nerve Activity Contributes to Central Artery Stiffness in Young and Middle-Age/Older Adults. *Hypertension* 73, 1025–1035. doi: 10.1161/HYPERTENSIONAHA.118.12462
- Huang, S.-S., He, S.-L., and Zhang, Y.-M. (2016). The effects of telmisartan on the nuclear factor of activated T lymphocytes signalling pathway in hypertensive patients. *J. Renin Angiotensin Aldosterone Syst.* 17:1470320316655005. doi: 10.1177/1470320316655005
- Hughes, W. E., Zinkevich, N., Guterman, D. D., and Beyer, A. M. (2021b). Hypertension preserves the magnitude of microvascular flow-mediated dilation following transient elevation in intraluminal pressure. *Physiol. Rep.* 9:e14507. doi: 10.14814/phy2.14507
- Hughes, W. E., Chabowski, D. S., Ait-Aissa, K., Fetterman, J. L., Hockenberry, J., Beyer, A. M., et al. (2021a). Critical Interaction Between Telomerase and Autophagy in Mediating Flow-Induced Human Arteriolar Vasodilation. *Arterioscler. Thromb. Vascul. Biol.* 41, 446–457. doi: 10.1161/ATVBAHA.120.314944
- Hurr, C., Patik, J. C., Kim, K., Christmas, K. M., and Brothers, R. M. (2018). Tempol augments the blunted cutaneous microvascular thermal reactivity in healthy young African Americans. *Exp. Physiol.* 103, 343–349. doi: 10.1113/EP086776
- Jacobi, J., Kristal, B., Chezar, J., Shaul, S. M., and Sela, S. (2005). Exogenous superoxide mediates pro-oxidative, proinflammatory, and procoagulatory changes in primary endothelial cell cultures. *Free Radic. Biol. Med.* 39, 1238–1248. doi: 10.1016/j.freeradbiomed.2005.06.010
- Ji, H., Kim, A., Ebinger, J. E., Niiranen, T. J., Claggett, B. L., Bairey Merz, C. N., et al. (2020). Sex Differences in Blood Pressure Trajectories Over the Life Course. *JAMA Cardiol.* 5, 19–26. doi: 10.1001/jamacardio.2019.5306
- Ji, W., Liu, S., Dai, J., Yang, T., Jiang, X., Duan, X., et al. (2014). Hydrogen sulfide defends against the cardiovascular risk of Nw-nitro-L-argininemethyl ester-induced hypertension in rats via the nitric oxide/endothelial nitric oxide synthase pathway. *Chin. Med. J.* 127, 3751–3757.
- Johnson, E. N., Lee, Y. M., Sander, T. L., Rabkin, E., Schoen, F. J., Kaushal, S., et al. (2003). NFATc1 mediates vascular endothelial growth factor-induced proliferation of human pulmonary valve endothelial cells. *J. Biol. Chem.* 278, 1686–1692. doi: 10.1074/jbc.M210250200
- Junqueira, C. L. C., Magalhães, M. E. C., Brandão, A. A., Ferreira, E., Junqueira, A. S. M., Neto, J. F. N., et al. (2018). Evaluation of endothelial function by VOP and inflammatory biomarkers in patients with arterial hypertension. *J. Hum. Hypertens.* 32, 105–113. doi: 10.1038/s41371-017-0024-z
- Juonala, M., Viikari, J. S., Laitinen, T., Marniemi, J., Helenius, H., Rönnemaa, T., et al. (2004). Interrelations between brachial endothelial function and carotid intima-media thickness in young adults: the cardiovascular risk in young Finns study. *Circulation* 110, 2918–2923. doi: 10.1161/01.CIR.0000147540.88559.00
- Kadlec, A. O., Chabowski, D. S., Ait-Aissa, K., Hockenberry, J. C., Otterson, M. F., Durand, M. J., et al. (2017). PGC-1alpha (Peroxisome Proliferator-Activated Receptor gamma Coactivator 1-alpha) Overexpression in Coronary Artery Disease Recruits NO and Hydrogen Peroxide During Flow-Mediated Dilatation and Protects Against Increased Intraluminal Pressure. *Hypertension* 70, 166–173. doi: 10.1161/HYPERTENSIONAHA.117.09289
- Karbach, S., Wenzel, P., Waisman, A., Munzel, T., and Daiber, A. (2014). eNOS uncoupling in cardiovascular diseases—the role of oxidative stress and inflammation. *Curr. Pharm. Des.* 20, 3579–3594. doi: 10.2174/1381612811319660748
- Katulka, E. K., Hirt, A. E., Kirkman, D. L., Edwards, D. G., and Witman, M. A. H. (2019). Altered vascular function in chronic kidney disease: evidence from passive leg movement. *Physiol. Rep.* 7:e14075. doi: 10.14814/phy2.14075
- Keir, D. A., Badrov, M. B., Tomlinson, G., Notarius, C. F., Kimmerly, D. S., Millar, P. J., et al. (2020). Influence of Sex and Age on Muscle Sympathetic

- Nerve Activity of Healthy Normotensive Adults. *Hypertension* 76, 997–1005. doi: 10.1161/HYPERTENSIONAHA.120.15208
- Kiani, A., Rao, A., and Aramburu, J. (2000). Manipulating immune responses with immunosuppressive agents that target NFAT. *Immunity* 12, 359–372. doi: 10.1016/S1074-7613(00)80188-0
- Kirkman, D. L., Robinson, A. T., Rossman, M. J., Seals, D. R., and Edwards, D. G. (2021). Mitochondrial contributions to vascular endothelial dysfunction, arterial stiffness, and cardiovascular diseases. *Am. J. Physiol. Heart Circ. Physiol.* 320, H2080–H2100. doi: 10.1152/ajpheart.00917.2020
- Kopp, E., and Ghosh, S. (1994). Inhibition of NF- κ B by sodium salicylate and aspirin. *Science* 265, 956–959. doi: 10.1126/science.8052854
- Lee, K. S., Kim, J., Kwak, S. N., Lee, K. S., Lee, D. K., Ha, K. S., et al. (2014). Functional role of NF- κ B in expression of human endothelial nitric oxide synthase. *Biochem. Biophys. Res. Communicat.* 448, 101–107. doi: 10.1016/j.bbrc.2014.04.079
- Lewington, S., Clarke, R., Qizilbash, N., Peto, R., and Collins, R. (2002). Age-specific relevance of usual blood pressure to vascular mortality: a meta-analysis of individual data for one million adults in 61 prospective studies. *Lancet* 360, 1903–1913. doi: 10.1016/S0140-6736(02)11911-8
- Li, L., Sapkota, M., Kim, S. W., and Soh, Y. (2015). Herbacetin inhibits inducible nitric oxide synthase via JNK and nuclear factor- κ B in LPS-stimulated RAW264.7 cells. *Eur. J. Pharmacol.* 765, 115–123. doi: 10.1016/j.ejphar.2015.08.032
- Lindstedt, I. H., Edvinsson, M. L., and Edvinsson, L. (2006). Reduced responsiveness of cutaneous microcirculation in essential hypertension—a pilot study. *Blood Press.* 15, 275–280. doi: 10.1080/08037050600996586
- Lipskaia, L., Pourci, M. L., Delomenie, C., Combettes, L., Goudouneche, D., Paul, J. L., et al. (2003). Phosphatidylinositol 3-kinase and calcium-activated transcription pathways are required for VLDL-induced smooth muscle cell proliferation. *Circ. Res.* 92, 1115–1122. doi: 10.1161/01.RES.0000074880.25540. D0
- Marin, T., Gongol, B., Chen, Z., Woo, B., Subramaniam, S., Chien, S., et al. (2013). Mechanosensitive microRNAs—role in endothelial responses to shear stress and redox state. *Free Radic. Biol. Med.* 64, 61–68. doi: 10.1016/j.freeradbiomed.2013.05.034
- Martens, C. R., Denman, B. A., Mazzo, M. R., Armstrong, M. L., Reisdorff, N., McQueen, M. B., et al. (2018). Chronic nicotinamide riboside supplementation is well-tolerated and elevates NAD(+) in healthy middle-aged and older adults. *Nat. Commun.* 9:1286. doi: 10.1038/s41467-018-03421-7
- Martens, C. R., Kirkman, D. L., and Edwards, D. G. (2016). The Vascular Endothelium in Chronic Kidney Disease: A Novel Target for Aerobic Exercise. *Exerc. Sport Sci. Rev.* 44, 12–19. doi: 10.1249/JES.0000000000000065
- Matthews, E. L., Greaney, J. L., and Wenner, M. M. (2017). Rapid onset pressor response to exercise in young women with a family history of hypertension. *Exp. Physiol.* 102, 1092–1099. doi: 10.1113/EP086466
- McGowan, C. L., Levy, A. S., Millar, P. J., Guzman, J. C., Morillo, C. A., McCartney, N., et al. (2006). Acute vascular responses to isometric handgrip exercise and effects of training in persons medicated for hypertension. *Am. J. Physiol. Heart Circ. Physiol.* 291, H1797–H1802. doi: 10.1152/ajpheart.01113.2005
- Mi, S., Zhang, J., Zhang, W., and Huang, R. S. (2013). Circulating microRNAs as biomarkers for inflammatory diseases. *Microrna* 2, 63–71. doi: 10.2174/221536611302010007
- Migrino, R. Q., Truran, S., Guterman, D. D., Franco, D. A., Bright, M., Schlundt, B., et al. (2011). Human microvascular dysfunction and apoptotic injury induced by AL amyloidosis light chain proteins. *Am. J. Physiol. Heart Circ. Physiol.* 301, H2305–H2312. doi: 10.1152/ajpheart.00503.2011
- Mills, K. F., Yoshida, S., Stein, L. R., Grozio, A., Kubota, S., Sasaki, Y., et al. (2016). Long-Term Administration of Nicotinamide Mononucleotide Mitigates Age-Associated Physiological Decline in Mice. *Cell Metab.* 24, 795–806. doi: 10.1016/j.cmet.2016.09.013
- Nieves-Cintrón, M., Amberg, G. C., Nichols, C. B., Molkentin, J. D., and Santana, L. F. (2007). Activation of NFATc3 down-regulates the beta1 subunit of large conductance, calcium-activated K+ channels in arterial smooth muscle and contributes to hypertension. *J. Biol. Chem.* 282, 3231–3240. doi: 10.1074/jbc.M608822200
- Nilsson, J., Nilsson, L. M., Chen, Y. W., Molkentin, J. D., Erlinge, D., and Gomez, M. F. (2006). High glucose activates nuclear factor of activated T cells in native vascular smooth muscle. *Arterioscler. Thromb. Vascul. Biol.* 26, 794–800. doi: 10.1161/01.ATV.0000209513.00765.13
- Nilsson-Berglund, L. M., Zetterqvist, A. V., Nilsson-Ohman, J., Sigvardsson, M., Gonzalez Bosc, L. V., Smith, M. L., et al. (2010). Nuclear factor of activated T cells regulates osteopontin expression in arterial smooth muscle in response to diabetes-induced hyperglycemia. *Arterioscler. Thromb. Vascul. Biol.* 30, 218–224. doi: 10.1161/ATVBAHA.109.199299
- Njock, M. S., and Fish, J. E. (2017). Endothelial miRNAs as Cellular Messengers in Cardiometabolic Diseases. *Trends Endocrinol. Metab.* 28, 237–246. doi: 10.1016/j.tem.2016.11.009
- Norata, G. D., Grigore, L., Raselli, S., Redaelli, L., Hamsten, A., Maggi, F., et al. (2007). Post-prandial endothelial dysfunction in hypertriglyceridemic subjects: molecular mechanisms and gene expression studies. *Atherosclerosis* 193, 321–327. doi: 10.1016/j.atherosclerosis.2006.09.015
- Nowroozpoor, A., Guterman, D., and Safdar, B. (2021). Is microvascular dysfunction a systemic disorder with common biomarkers found in the heart, brain, and kidneys? - A scoping review. *Microvasc. Res.* 134:104123. doi: 10.1016/j.mvr.2020.104123
- On, Y. K., Kim, C. H., Sohn, D. W., Oh, B. H., Lee, M. M., Park, Y. B., et al. (2002). Improvement of endothelial function by amlodipine and vitamin C in essential hypertension. *Korean J. Intern. Med.* 17, 131–137. doi: 10.3904/kjim.2002.1.7.131
- Orr, A. W., Lee, M. Y., Lemmon, J. A., Yurdagul, A., Gomez, M. F., Bortz, P. D. S., et al. (2009). Molecular Mechanisms of Collagen Isootype-Specific Modulation of Smooth Muscle Cell Phenotype. *Arterioscl. Throm. Vas.* 29, 225–231. doi: 10.1161/ATVBAHA.108.178749
- Ozemek, C., Hildreth, K. L., Groves, D. W., and Moreau, K. L. (2016). Acute ascorbic acid infusion increases left ventricular diastolic function in postmenopausal women. *Maturitas* 92, 154–161. doi: 10.1016/j.maturitas.2016.08.007
- Pang, X., and Sun, N. L. (2009). Calcineurin-NFAT signaling is involved in phenylephrine-induced vascular smooth muscle cell proliferation. *Acta Pharmacol. Sin.* 30, 537–544. doi: 10.1038/aps.2009.28
- Park, S. Y., Kwon, O. S., Andtbacka, R. H. I., Hyngstrom, J. R., Reese, V., Murphy, M. P., et al. (2018). Age-related endothelial dysfunction in human skeletal muscle feed arteries: the role of free radicals derived from mitochondria in the vasculature. *Acta physiol.* 222:12893. doi: 10.1111/apha.12893
- Phillips, S. A., Hatoum, O. A., and Guterman, D. D. (2007). The mechanism of flow-induced dilation in human adipose arterioles involves hydrogen peroxide during CAD. *Am. J. Physiol. Heart Circ. Physiol.* 292, H93–H100. doi: 10.1152/ajpheart.00819.2006
- Pierce, G. L., Lesniewski, L. A., Lawson, B. R., Beske, S. D., and Seals, D. R. (2009). Nuclear factor- $\{\kappa\}B$ activation contributes to vascular endothelial dysfunction via oxidative stress in overweight/obese middle-aged and older humans. *Circulation* 119, 1284–1292. doi: 10.1161/CIRCULATIONAHA.108.804294
- Pierce, J. W., Read, M. A., Ding, H., Luscinskas, F. W., and Collins, T. (1996). Salicylates inhibit 1 κ B-alpha phosphorylation, endothelial-leukocyte adhesion molecule expression, and neutrophil transmigration. *J. Immunol.* 156, 3961–3969.
- Pohlmeus, D. J., and Lefer, D. J. (2014). Emergence of hydrogen sulfide as an endogenous gaseous signaling molecule in cardiovascular disease. *Circ. Res.* 114, 730–737. doi: 10.1161/CIRCRESAHA.114.300505
- Ratchford, S. M., Clifton, H. L., Gifford, J. R., LaSalle, D. T., Thurston, T. S., Bunsawat, K., et al. (2019a). Impact of acute antioxidant administration on inflammation and vascular function in heart failure with preserved ejection fraction. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 317, R607–R614. doi: 10.1152/ajpregu.00184.2019
- Ratchford, S. M., Broxterman, R. M., La Salle, D. T., Kwon, O. S., Park, S. Y., Hopkins, P. N., et al. (2019b). Salt restriction lowers blood pressure at rest and during exercise without altering peripheral hemodynamics in hypertensive individuals. *Am. J. Physiol. Heart Circ. Physiol.* 317, H1194–H1202.
- Richards, J. C., Creelius, A. R., Larson, D. G., and Dinrenno, F. A. (2015). Acute ascorbic acid ingestion increases skeletal muscle blood flow and oxygen

- consumption via local vasodilation during graded handgrip exercise in older adults. *Am. J. Physiol. Heart Circ. Physiol.* 309, H360–H368.
- Risbano, M. G., and Gladwin, M. T. (2013). Therapeutics targeting of dysregulated redox equilibrium and endothelial dysfunction. *Handb. Exp. Pharmacol.* 218, 315–349. doi: 10.1007/978-3-662-45805-1_13
- Rossman, M. J., Santos-Parker, J. R., Steward, C. A. C., Bispham, N. Z., Cuevas, L. M., Rosenberg, H. L., et al. (2018). Chronic Supplementation With a Mitochondrial Antioxidant (MitoQ) Improves Vascular Function in Healthy Older Adults. *Hypertension* 71, 1056–1063. doi: 10.1161/HYPERTENSIONAHA.117.10787
- Sapp, R. M., Chesney, C. A., Springer, C. B., Laskowski, M. R., Singer, D. B., Eagan, L. E., et al. (2021). Race-specific changes in endothelial inflammation and microRNA in response to an acute inflammatory stimulus. *Am. J. Physiol. Heart Circ. Physiol.* 320, H2371–H2384. doi: 10.1152/ajpheart.00991.2020
- Sapp, R. M., Shill, D. D., Roth, S. M., and Hagberg, J. M. (2017). Circulating microRNAs in acute and chronic exercise: more than mere biomarkers. *J. Appl. Physiol.* 122, 702–717. doi: 10.1152/japplphysiol.00982.2016
- Schmitz, B., Breulmann, F. L., Jubran, B., Rolfs, F., Thorwesten, L., Kruger, M., et al. (2019). A three-step approach identifies novel shear stress-sensitive endothelial microRNAs involved in vasculoprotective effects of high-intensity interval training (HIIT). *Oncotarget* 10, 3625–3640. doi: 10.18632/oncotarget.26944
- Selemidis, S., Dusting, G. J., Peshavariya, H., Kemp-Harper, B. K., and Drummond, G. R. (2007). Nitric oxide suppresses NADPH oxidase-dependent superoxide production by S-nitrosylation in human endothelial cells. *Cardiovasc. Res.* 75, 349–358. doi: 10.1016/j.cardiores.2007.03.030
- Shi, L., Liao, J., Liu, B., Zeng, F., and Zhang, L. (2015). Mechanisms and therapeutic potential of microRNAs in hypertension. *Drug Discov. Today* 20, 1188–1204. doi: 10.1016/j.drudis.2015.05.007
- Shiny, A., Regin, B., Mohan, V., and Balasubramanyam, M. (2016). Coordinated augmentation of NFAT and NOD signaling mediates proliferative VSMC phenotype switch under hyperinsulinemia. *Atherosclerosis* 246, 257–266. doi: 10.1016/j.atherosclerosis.2016.01.006
- Smith, C. J., Santhanam, L., and Alexander, L. M. (2013). Rho-Kinase activity and cutaneous vasoconstriction is upregulated in essential hypertensive humans. *Microvascul. Res.* 87, 58–64. doi: 10.1016/j.mvr.2013.02.005
- Smith, C. J., Santhanam, L., Bruning, R. S., Stanhewicz, A., Berkowitz, D. E., and Holowatz, L. A. (2011). Upregulation of inducible nitric oxide synthase contributes to attenuated cutaneous vasodilation in essential hypertensive humans. *Hypertension* 58, 935–942. doi: 10.1161/HYPERTENSIONAHA.111.178129
- Soudani, N., Ghantous, C. M., Farhat, Z., Shebaly, W. N., Zibara, K., and Zeidan, A. (2016). Calcineurin/NFAT Activation-Dependence of Leptin Synthesis and Vascular Growth in Response to Mechanical Stretch. *Front. Physiol.* 7:433. doi: 10.3389/fphys.2016.00433
- Sprick, J. D., Downey, R. M., Morison, D. L., Fonkoue, I. T., Li, Y., DaCosta, D., et al. (2019). Functional sympatholysis is impaired in end-stage renal disease. *Am. J. Physiol. Regul. Integr. Comp. Physiol.* 316, R504–R511. doi: 10.1152/ajpregu.00380.2018
- Stanhewicz, A. E., Alexander, L. M., and Kenney, W. L. (2015). Folic acid supplementation improves microvascular function in older adults through nitric oxide-dependent mechanisms. *Clin. Sci.* 129, 159–167. doi: 10.1042/CS20140821
- Stanhewicz, A. E., and Kenney, W. L. (2017). Role of folic acid in nitric oxide bioavailability and vascular endothelial function. *Nutrit. Rev.* 75, 61–70. doi: 10.1093/nutrit/nuw053
- Suzuki, E., Nishimatsu, H., Satonaka, H., Walsh, K., Goto, A., Omata, M., et al. (2002). Angiotensin II induces myocyte enhancer factor 2- and calcineurin/nuclear factor of activated T cell-dependent transcriptional activation in vascular myocytes. *Circ. Res.* 90, 1004–1011. doi: 10.1161/01.RES.0000017629.70769.CC
- Takase, B., Hamabe, A., Satomura, K., Akima, T., Uehata, A., Matsui, T., et al. (2006). Comparable prognostic value of vasodilator response to acetylcholine in brachial and coronary arteries for predicting long-term cardiovascular events in suspected coronary artery disease. *Circ. J.* 70, 49–56. doi: 10.1253/circj.70.49
- Trinity, J. D., Wray, D. W., Witman, M. A., Layec, G., Barrett-O'Keefe, Z., Ives, S. J., et al. (2016). Ascorbic acid improves brachial artery vasodilation during progressive handgrip exercise in the elderly through a nitric oxide-mediated mechanism. *Am. J. Physiol. Heart Circ. Physiol.* 310, H765–H774. doi: 10.1152/ajpheart.00817.2015
- Virani, S. S., Alonso, A., Aparicio, H. J., Benjamin, E. J., Bittencourt, M. S., Callaway, C. W., et al. (2021). Heart Disease and Stroke Statistics-2021 Update: A Report From the American Heart Association. *Circulation* 143, e254–e743. doi: 10.1161/CIR.0000000000000950
- Wadley, A. J., Veldhuijen van Zanten, J. J., and Aldred, S. (2013). The interactions of oxidative stress and inflammation with vascular dysfunction in ageing: the vascular health triad. *Age* 35, 705–718. doi: 10.1007/s11357-012-9402-1
- Walker, A. E., Kaplon, R. E., Pierce, G. L., Nowlan, M. J., and Seals, D. R. (2014). Prevention of age-related endothelial dysfunction by habitual aerobic exercise in healthy humans: possible role of nuclear factor κB. *Clin. Sci.* 127, 645–654. doi: 10.1042/CS20140030
- Wang, Q., Li, D., Han, Y., Ding, X., Xu, T., and Tang, B. (2017). MicroRNA-146 protects A549 and H1975 cells from LPS-induced apoptosis and inflammation injury. *J. Biosci.* 42, 637–645. doi: 10.1007/s12038-017-9715-4
- Wang, Y., Hu, J., Liu, J., Geng, Z., Tao, Y., Zheng, F., et al. (2020). The role of Ca(2+)/NFAT in Dysfunction and Inflammation of Human Coronary Endothelial Cells induced by Sera from patients with Kawasaki disease. *Sci. Rep.* 10:4706. doi: 10.1038/s41598-020-61667-y
- Weng, J., Wang, C., Zhong, W., Li, B., Wang, Z., Shao, C., et al. (2017). Activation of CD137 Signaling Promotes Angiogenesis in Atherosclerosis via Modulating Endothelial Smad1/5-NFATc1 Pathway. *J. Am. Heart Assoc.* 6:e004756. doi: 10.1161/JAHA.116.004756
- Whelton, P. K., Carey, R. M., Aronow, W. S., Casey, D. E. Jr., Collins, K. J., Dennison Himmelfarb, C., et al. (2018). 2017 ACC/AHA/AAPA/ABC/ACPM/AGS/APhA/ASH/ASPC/NMA/PCNA Guideline for the Prevention, Detection, Evaluation, and Management of High Blood Pressure in Adults: A Report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. *J. Am. Coll. Cardiol.* 71, e127–e248. doi: 10.1161/HYP.0000000000000076
- Wray, D. W., Nishiyama, S. K., Harris, R. A., Zhao, J., McDaniel, J., Fjeldstad, A. S., et al. (2012). Acute reversal of endothelial dysfunction in the elderly after antioxidant consumption. *Hypertension* 59, 818–824. doi: 10.1161/HYPERTENSIONAHA.111.189456
- Wu, F., Tyml, K., and Wilson, J. X. (2008). iNOS expression requires NADPH oxidase-dependent redox signaling in microvascular endothelial cells. *J. Cell Physiol.* 217, 207–214. doi: 10.1002/jcp.21495
- Xue, H., Zhou, S., Xiao, L., Guo, Q., Liu, S., and Wu, Y. (2015). Hydrogen sulfide improves the endothelial dysfunction in renovascular hypertensive rats. *Physiol. Res.* 64, 663–672. doi: 10.3354/physiolres.932848
- Yamakuchi, M. (2012). MicroRNA Regulation of SIRT1. *Front. Physiol.* 3:68. doi: 10.3389/fphys.2012.00068
- Yamakuchi, M., and Hashiguchi, T. (2018). Endothelial Cell Aging: How miRNAs Contribute? *J. Clin. Med.* 7:170. doi: 10.3390/jcm7070170
- Yan, J., Yin, Y., Zhong, W., Wang, C., and Wang, Z. (2015). CD137 Regulates NFATc1 Expression in Mouse VSMCs through TRAF6/NF-κB p65 Signaling Pathway. *Meditat. Inflamm.* 2015:639780. doi: 10.1155/2015/639780
- Yang, P., Liu, Y. F., Yang, L., Wei, Q., and Zeng, H. (2010). Mechanism and clinical significance of the prothrombotic state in patients with essential hypertension. *Clin. Cardiol.* 33, E81–E86. doi: 10.1002/clc.20719
- Yoshino, J., Mills, K. F., Yoon, M. J., and Imai, S. (2011). Nicotinamide mononucleotide, a key NAD(+) intermediate, treats the pathophysiology of diet- and age-induced diabetes in mice. *Cell Metab.* 14, 528–536. doi: 10.1016/j.cmet.2011.08.014
- Zelko, I. N., Mariani, T. J., and Folz, R. J. (2002). Superoxide dismutase multigene family: a comparison of the CuZn-SOD (SOD1), Mn-SOD (SOD2), and EC-SOD (SOD3) gene structures, evolution, and expression. *Free Radic. Biol. Med.* 33, 337–349. doi: 10.1016/S0891-5849(02)00905-X
- Zetterqvist, A. V., Berglund, L. M., Blanco, F., Garcia-Vaz, E., Wigren, M., Duner, P., et al. (2014). Inhibition of nuclear factor of activated T-cells (NFAT) suppresses accelerated atherosclerosis in diabetic mice. *PLoS One* 8:e65020. doi: 10.1371/journal.pone.0065020

- Zetterqvist, A. V., Blanco, F., Öhman, J., Kotova, O., Berglund, L. M., de Frutos Garcia, S., et al. (2015). Nuclear factor of activated T cells is activated in the endothelium of retinal microvessels in diabetic mice. *J. Diabet. Res.* 2015:428473. doi: 10.1155/2015/428473
- Zhang, L., Chen, J., He, Q., Chao, Z., Li, X., and Chen, M. (2019). MicroRNA-217 is involved in the progression of atherosclerosis through regulating inflammatory responses by targeting sirtuin 1. *Mol. Med. Rep.* 20, 3182–3190. doi: 10.3892/mmr.2019.10581
- Zhao, W., Zhang, J., Lu, Y., and Wang, R. (2001). The vasorelaxant effect of H(2)S as a novel endogenous gaseous K(ATP) channel opener. *EMBO J.* 20, 6008–6016. doi: 10.1093/emboj/20.21.6008
- Zhong, L., Simard, M. J., and Huot, J. (2018). Endothelial microRNAs regulating the NF-kappaB pathway and cell adhesion molecules during inflammation. *FASEB J.* 32, 4070–4084. doi: 10.1096/fj.201701536R

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2021 Ranadive, Dillon, Mascone and Alexander. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.