Yesao special issue

IJAO The International Journal of Artificial Organs

Effects of high-frequency mechanical stimuli on flow related vascular cell biology

The International Journal of Artificial Organs
1–12

DOI: 10.1177/03913988241268105
journals.sagepub.com/home/jao
Sage

Elena Carrara[1], Luca Soliveri[1], Sofia Poloni[2], Michela Bozzetto[1] and Chiara Emma Campiglio[3]

Abstract

Mechanical forces related to blood pressure and flow patterns play a crucial role in vascular homeostasis. Perturbations in vascular stresses and strain resulting from changes in hemodynamic may occur in pathological conditions, leading to vascular dysfunction as well as in vascular prosthesis, arteriovenous shunt for hemodialysis and in mechanical circulation support. Turbulent-like blood flows can induce high-frequency vibrations of the vessel wall, and this stimulus has recently gained attention as potential contributors to vascular pathologies, such as development of intimal hyperplasia in arteriovenous fistula for hemodialysis. However, the biological response of vascular cells to this stimulus remains incompletely understood. This review provides an analysis of the existing literature concerning the impact of high-frequency stimuli on vascular cell morphology, function, and gene expression. Morphological and functional investigations reveal that vascular cells stimulated at frequencies higher than the normal heart rate exhibit alterations in cell shape, alignment, and proliferation, potentially leading to vessel remodeling. Furthermore, vibrations modulate endothelial and smooth muscle cells gene expression, affecting pathways related to inflammation, oxidative stress, and muscle hypertrophy. Understanding the effects of high-frequency vibrations on vascular cells is essential for unraveling the mechanisms underlying vascular diseases and identifying potential therapeutic targets. Nevertheless, there are still gaps in our understanding of the molecular pathways governing these cellular responses. Further research is necessary to elucidate these mechanisms and their therapeutic implications for vascular diseases.

Keywords

Vascular cells, high-frequency, vibration, tissue remodeling, mechanobiology

Date received: 30 April 2024; accepted: 12 July 2024

Introduction

Non-physiological stress and strain on vascular wall

Blood vessels are constantly exposed to two hemodynamic stimuli due to the pulsatile nature of blood pressure and flow: the wall shear stress (WSS), which is the frictional force per unit area exerted by the blood flow parallel to the vessel wall, and the intraluminal pressure, which stretches the vessel wall perpendicularly, promoting circumferential strain.[1,2] Vascular cells sense and respond to these mechanical stimuli to maintain the integrity of the vasculature itself and to enable appropriate adaptations.[3] Despite vascular endothelial cells (ECs) and vascular smooth muscle cells (SMCs) are exposed to both types of mechanical cues, the shear stress resulting from blood flow is acting primarily on ECs of the intima, whereas SMCs of the media are primarily subjected and sense changes in intramural stress.[1,3]

[1]Department of Biomedical Engineering, Istituto di Ricerche Farmacologiche Mario Negri IRCCS, Bergamo, Italy
[2]Department of Engineering and Applied Sciences, University of Bergamo, Dalmine, Italy
[3]Department of Management, Information and Production Engineering, University of Bergamo, Dalmine, Italy

Corresponding author:
Elena Carrara, Department of Biomedical Engineering, Istituto di Ricerche Farmacologiche Mario Negri IRCCS, via Stezzano, 87, Bergamo 24126, Italy.
Email: elena.carrara@marionegri.it

In this context, several molecular pathways are known to be involved in the onset of cellular responses to the above-mentioned mechanical forces,[1] which result in the regulation of intracellular signaling, gene expression, and protein expression that maintain the physiological function of blood vessels. In physiological conditions, the biological response varies with changes in mechanical and chemical stimuli, maintaining a dynamic balance which allows to preserve vascular homeostasis.[4] However, when this balance is disturbed by non-physiological mechanical stimuli, cell signaling can lead to a responsive adaptation or damaging of the vessel wall, possibly contributing to diseases of the vasculature. In this regards, previous research has demonstrated that ECs exposed to unidirectional and elevated WSS exhibit a quiescent phenotype[5,6] while disturbed flow characterized by low and oscillatory WSS, typically found in bifurcation or branching regions, has been suggested to trigger a proliferative, pro-inflammatory, pro-oxidant ECs state and an impaired vascular tone regulation.[5] In literature, it is also reported that SMCs manifest a diverse array of phenotypes, based on the type of artery and the presence of disease. Additionally, different loads can promote various cytoplasmic signals and gene expression patterns that modulate both SMCs' structure and phenotype.[3] Typically, cyclic strains or stresses at levels that replicate physiological values result in maintaining physiological SMCs phenotypes. On the contrary, exposure to cyclic strains or stresses exceeding normal thresholds often triggers inflammatory pathways in SMCs.[7,8]

Perturbations in vascular stresses and strains caused by changes in blood pressure and hemodynamics may occur either under physiological conditions, such as during physical exercise, or pathological conditions related to vascular disease.[9] Changes in physiological vascular stresses develop also in arteriovenous shunt for vascular access in hemodialysis patients, as well as in artificial vascular graft and in some conditions also with mechanical circulatory support. Thus, over the past decades it has become evident that understanding how the local hemodynamic conditions affect the mechanotransduction of vascular cells is crucial for elucidating the underlying mechanisms of vascular homeostasis and disease.

Turbulent-like flow-induced vascular wall vibrations revealed through computational studies

Disturbed blood flow and "turbulence" in the vascular system is normally linked to pathological conditions. These flows feature rapid and seemingly random velocity and pressure fluctuations in time and space, with continuous energy transfer across scales. Such flows may share certain features with turbulence, but do not necessarily exhibit all of the well-known characteristics outlined by Tennekes and Lumley in 1972 or follow mathematical theories of high Reynolds number homogeneous and isotropic turbulence itself.[10]

Furthermore, the presence of flow instabilities leading to vascular pathologies and adverse vascular remodeling is well-documented in various vascular districts. For instance, adopting medical image-based computational fluid dynamics (CFD) from magnetic resonance imaging (MRI) or computed tomography (CT) scans, with boundary conditions obtained from literature or patient-specific Doppler ultrasound, turbulent-like flow phenotypes have been documented in cerebral aneurysms,[11] as well as in carotid syphons[12] where atherosclerotic plaques commonly occur. Additionally, transitional flow has been found in the venous segment of hemodialysis arteriovenous fistula (AVF),[13,14] a vascular region usually associated to neointimal hyperplasia,[15] which is a fibrotic-muscular thickening of the vessel wall caused by the migration of smooth muscle cells into the intimal layer of the vessel.[16,17]

Such turbulent-like flow can induce high-frequency vibrations in the vascular wall. Recent studies from our group[18] showed, through a fluid-structure interaction (FSI) approach, that transitional flow-induced high frequency pressure fluctuations caused the walls in the AVF vein to vibrate at frequencies up to hundreds of Hz. Moreover, these vibration amplitudes were found to be predominant at the inner curvature of the cephalic vein, the region where stenotic lesions typically develop. Furthermore, recent FSI studies have uncovered the presence of vascular wall vibrations also within cerebral aneurysms,[10,19,20] unraveling broad-band, random vibrations which show similar characteristics with clinically observed bruits.[21,22]

All these evidence from "in silico" studies suggest that high-frequency vibrations may play a role in vascular mechanobiology across different vascular regions, highlighting the need for further exploration into this phenomenon.

Vascular wall vibrations in the onset of cardiovascular disease

Investigating the impact of high-frequency vibrations on the cardiovascular system has roots dating back to the latter half of the 20th century. During this period, experimental setups and animal clinical models were developed to explore thrills, bruits, and high-frequency vibrations, suggesting a potential link to the onset of vascular diseases. Already in 1963, Roach.[23] observed that the presence of a recordable bruits was a common feature in various diseases characterized by vascular stenosis. Wang et al.[24] found that patients with occlusive coronary arteries, where turbulent flow fluctuations were present, exhibited a typical increase in high frequencies that matched with acoustic signals recorded by a chest microphone. In the same period, other experiments[25–27] documented vascular wall vibrations that might contribute to the pathogenesis of structural fatigue affecting the vasculature subjected to

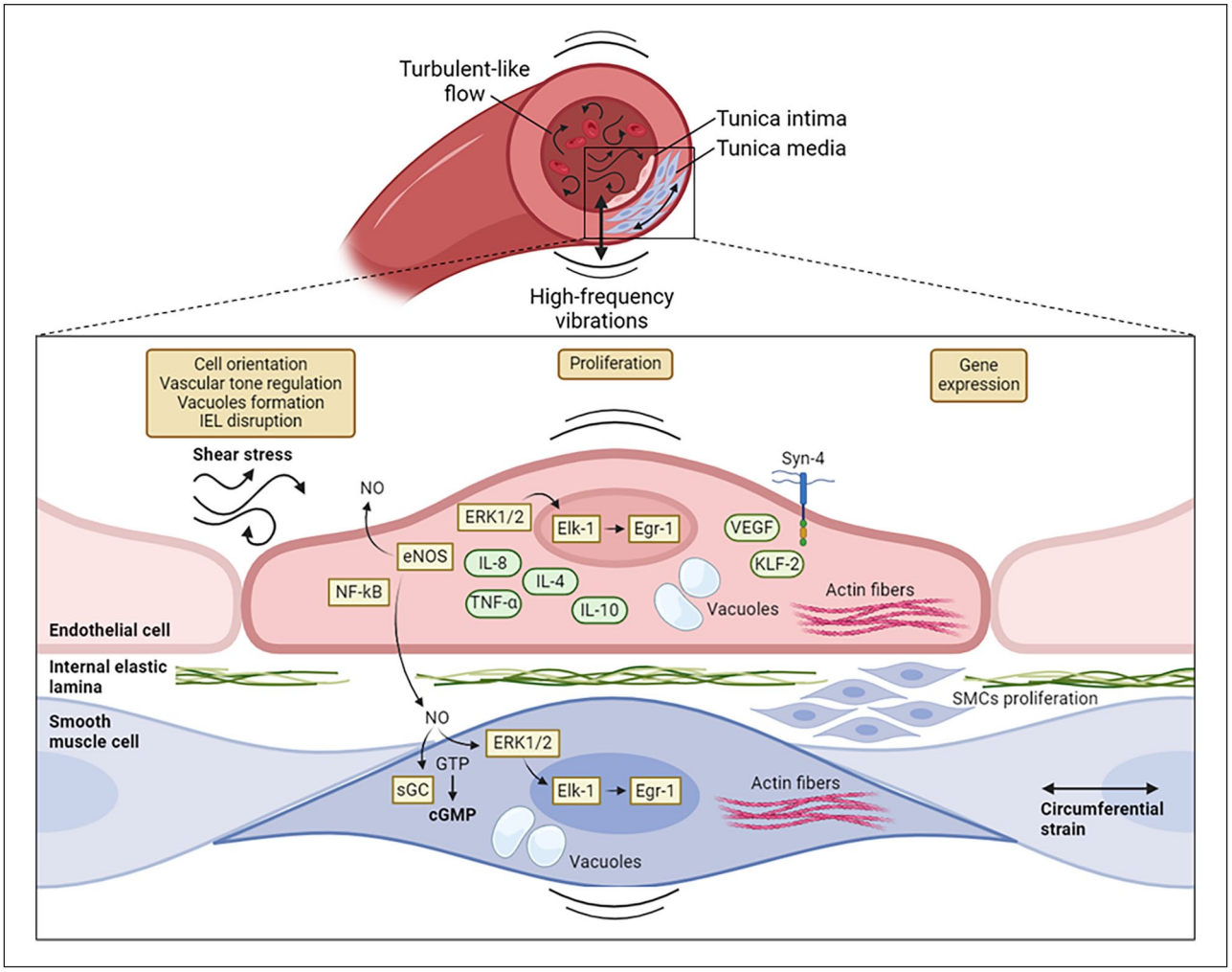

Figure 1. Biological response of vascular cells to flow-induced high-frequency vibrations. The schematic diagram reports some of the morphological, functional, and gene expression changes observed in endothelial (ECs) and smooth muscle cells (SMCs) exposed to stimuli at high-frequencies. Morphological changes include actin fibers alignment and cell orientation, vascular tone regulation, formation of double-membrane limited vacuoles, and disruption of the internal elastic lamina (IEL). Vibrations induce the proliferation of smooth muscle cells leading to a thickening of the intimal layer and promote in ECs and SMCs the expression of factors and the activation of pathways related to inflammation, oxidative stress and vascular dysfunction, including extracellular signal-regulated kinase 1 and 2 (ERK1/2), endothelial nitric-oxide synthase (eNOS), nuclear factor-kB (NF-kB), soluble guanylate cyclase (sGC), Krüppel-like factor 2 (KLF-2), vascular endothelial growth factor (VEGF), tumour necrosis factor alpha (TNF-α), interleukin-8 (IL-8), interleukin-4 (IL-4), and interleukin-10 (IL-10). Early growth response 1 (Egr-1). This figure was created using Biorender.com

non-physiological stimulus. Then, the relationship between bruits and pressure fluctuations in the post-stenotic region was numerically investigated in idealized vessels.[28,29] Also, recent studies on carotid artery bifurcation[30] and cerebral aneurysm[31] reported turbulent flow-induced high-frequency vibrations. Therefore, based on the reported rationale, understanding how the frequency of blood flow- and pressure-induced vessel wall displacement impacts on vascular function is essential for elucidating the underlying mechanisms of vascular diseases. Thus, the aim of this review is to explore the existing literature regarding the effect of a high-frequency vibration on the behavior of vascular cells in terms of morphological, functional, and mechanobiological changes. We define as high-frequency vibrations, frequencies exceeding that induced by the normal heart rate, corresponding approximately to 1–2 Hz in humans and to 6 and 9 Hz in rats and mice, respectively (Figure 1).[32]

Impact of high-frequency stimulation on vascular cells morphology, functions, and mechanobiology

Cellular mechanotransduction is an important biological process in living organisms, deeply investigated to assess the impact of biological mechanical cues such as the

circumferential stretching force, the fluid shear stress and the extracellular matrix (ECM) stiffness on multiple pathophysiological processes. Signals are sensed by the ion channels and receptors on the cell membrane and affect cell-matrix communications and signal transduction, triggering changes in cytoskeleton structure and downstream signaling cascades which regulate several processes including cell differentiation, adhesion, migration, proliferation, secretion of factors, and ECM generation.[33]

Many signaling pathways have been identified as involved in cell responding to mechanical stimuli. A large body of evidence is reported regarding the role of vascular endothelial growth factor (VEGF), Notch, platelet-derived growth factor (PDGF), Krüppel-like factor 2 (Klf2), endothelial nitric-oxide synthase (eNOS), endothelin, Rho family signaling molecules, mitogen-activated protein kinase (MAPK) signaling pathway, nuclear factor-kB (NF-κB) signaling pathway, and GTPase signaling pathway.[34,35] Great attention was given to mechanosensors including integrins, the glycocalyx, primary cilia, G protein-coupled receptors, and ion channels[36–43] such as Piezo1.[44] In response that is, to fluid shear stress, several mechanosensory complexes are also activated, including vascular endothelial cell cadherin (VE-cadherin), VEGF receptor 2 (VEGFR2), and platelet endothelial cell adhesion molecule (PECAM-1).[41] Furthermore, activation of extracellular signal-regulated kinase 1 and 2 (ERK1/2) cascades is reported.[45]

With regard to cellular responses to cues, several investigations have explored the connection between mechanical signal transduction and diseases. That being said, the study of vibrations of the blood vessel wall promoted by pressure fluctuations recently acquired interest, since a correlation with some vascular pathologies has been observed.[19,20,23,31] Based on these studies, high-frequency vibrations appear to impact vascular cell functions, suggesting the role of a mechanobiological process that links high-frequency stresses within the vessel wall to adverse vascular remodeling. In the next paragraphs, we report the available evidence in the literature concerning the effect of high-frequency stimuli on the biology of vascular cells.

Morphological changes in vascular cells stimulated by high-frequency stimuli

Over the years, the potential of high-frequency stimuli in inducing cell morphological changes has been studied by exposing cultured cells or vessel segments to mechanical loadings such as vibration or cyclic stretching. It is reasonable that cells sense a cyclic circumferential strain when they are subjected to a vertical vibration, since their body is stretched perpendicularly to the axis of their displacement. That being said, this stress might play a role in the effect of vibrations on vascular cells.

Several in vivo studies investigated the impact of this stimulus on vascular histology. Krajnak et al.[46] explored the physiological effects of vibrations on the ventral tail artery of rats exposed to a 125 Hz-vibration. Their study revealed no apparent signs of trauma to the vascular smooth muscle or endothelial cell layer in arteries collected from both stimulated and unstimulated animals. However, arteries from vibrated rat tails exhibited larger involutions in the internal elastic lamina (IEL), indicative of vessel constriction, and compression of endothelial cells.

Furthermore, in 2002, Curry investigated the mechanism of vibration injury by exposing rat tails to a 60 Hz vibration.[47] Electron microscopy revealed numerous arterial regions with loss and thinning of endothelial cells, along with activated platelets coating the exposed subendothelial tissue. Both ECs and SMCs contained double membrane-limited, swollen vacuoles similar to those formed under massive vasoconstriction induced by direct norepinephrine application.[48] This similarity suggests the vasoconstrictive potential of vibrations. Additionally, the study identified an increase in chondroitin sulfate proteoglycan, which normally precedes smooth muscle migration,[49] in the extracellular matrix between SMCs. Since the denudation of the endothelial barrier triggers platelet adherence and their release of factors leading to smooth muscle proliferation and stenosis, together with factors that degrade the internal elastic lamina, Curry hypothesized that a similar scenario may occur when vibration disrupts the endothelium of the tail artery, resulting in degradation of the IEL. If this hypothesis is correct, prolonged vibrations should result in smooth muscle overgrowth and eventually to vascular occlusion.

Much evidence shows that mechanical stresses can also modulate cell orientation and actin fibers alignment. For instance, cyclic strain at low frequencies has been observed to induce vascular cells to align most frequently approximately 90° away from the direction of stretch.[50–53]

In this regard, the orientation and structural network of the vascular SMCs layers that build up the arterial wall are very important for maintaining its mechanical strength and function and also for providing the mechanical compliance required for pulsatile blood flow.[54–56] The stress loading induces vascular smooth muscle cells to align in a frequency-dependent way,[50] and their response to cyclic mechanical loading appears to accelerate with higher frequencies.[51] However, this phenomenon was assessed only for frequencies lower than 2 Hz, indicating a need for further data on the outcomes of higher-frequency stimulation of these cells. Furthermore, ECs exhibit a greater degree of alignment in response to 1 Hz stretching compared to 0.1 Hz, resulting in a more attenuated effect, and to 0.01 Hz, which causes a complete lack of stress fiber orientation. Again, despite interesting studies on the response of these

cells to low frequencies, information about the impact of high-frequencies is not present in the literature.[52] Human fibroblasts subjected to cyclic stress elongate along the direction perpendicular to the mechanical loading, and the timing of the cytoskeleton orientation is influenced by cell density and stimulus frequency. For subconfluent cells, the time needed for the alignment decreases as the frequency of stretch loading raise from 0.1 Hz to 1 Hz. However, no further acceleration in alignment is observed when the stimulus is increased up to 20 Hz. Conversely, confluent cells exhibit a shorter reorientation time, which tends to decrease with increasing frequency of the applied stimulus. These findings suggest that cell-cell contacts mediated by cadherins may play a significant role in the cellular response to mechanical strain.[53]

Additionally, in a recent study by Mu et al., the impact of a stimulation with vibrations at a frequency of 31.5 Hz on the morphology of cultured ECs was assessed. They observed that in the absence of vibrations cells exhibited a flat, polygonal morphology, tightly packed in a paving stone pattern. However, this morphology was notably altered after mechanical stimulation, with cells acquiring a rounded morphology and a decreased distribution density.[57]

Overall, these studies highlight those different frequencies of mechanical stimuli may induce diverse cellular responses in different cell types. Vascular cells adjust their shape and orientation to endure the cues they experience, leading to the remodeling of the vessel wall which aims at the maintenance of the structure, function, and mechanical integrity of the vessel. However, despite the large number of studies, primarily investigating histological modifications of the vessel wall, the molecular mechanism by which vibration leads to these changes remains unknown. Moreover, consistent observation of the effect of vibrations within the range of 50–150 Hz is not yet present in the literature.

Functional changes in vascular cells stimulated by high-frequency stimuli

Mechanical signals exert significant influence over numerous essential cellular functions, impacting processes such as cell proliferation, differentiation, and migration, and consequently governing critical phenomena including bone and cartilage growth, wound healing, and angiogenesis.[58] However, many aspects of the biochemical transduction of some mechanical stresses like vibrations are not known in detail, yet a profound understanding of the way cells respond to them is important for basic biological science, in the vascular field in particular.

It has been reported that a normal response of biological tissues to repeated stress is an increase in proliferation.[59] Therefore, it is reasonable to hypothesize that exposure of vascular cells to high frequency stimulation may induce changes in cell growth and vessel remodeling. However, there is limited research available on this topic.

In vivo, prolonged exposure to vibrations triggers vascular impairment. Histological changes observed in the peripheral arteries of workers subjected to the frequent use of vibrating tools, who manifested Raynaud's phenomenon,[60] can be considered among the first evidence of the damaging potential of this mechanical stimulus. Raynaud's phenomenon is one of the main symptoms of the hand-arm vibration syndrome (HAVS), which is characterized by dysfunction of the peripheral vascular and sensorineural systems, and cause an exaggerated vasoconstriction, especially in response to cold temperature-exposure.[46,61]

Several studies investigated the role of wall vibrations in inducing vascular remodeling. Okada et al.[62] demonstrated experimentally the occurrence of intimal thickening in peripheral arteries of rats after 90 days of exposure to a local vibration at 60 Hz. The same group, some years later showed that the small arteries of hind legs of rats exposed for 30 days to vibrations at 30 and 480 Hz manifested a thicker intimal layer. The disruption of internal elastic lamina and focal cell proliferation and the formation of collagen and elastic fibres was also observed.[60] Further research[63] revealed an important reduction in the lumen diameter of ventral tail arteries in rats exposed to vibrations at 250 Hz, accompanied by a significant increase in vascular smooth muscle thickness. Conversely, stimulation at frequencies of 62.5 and 125 Hz induced morphological changes that were not statistically significant.

Additionally, Bittle[59] proposed a hypothesis trying to correlate the hyperproliferation of the intimal layer with vascular vibrations. She reported that the contractile myofilaments of vascular smooth muscle cells and their attachment to the plasma membrane as forming a lattice network. This network allows internal cellular structures, such as the nucleus and Golgi complex, to move differently from the surrounding cytoskeletal structure, resulting in abnormal stresses on cell structures. Such abnormal stress, along with localized stretching of the membrane due to relative motion of the myofilaments, may activate cellular signaling pathways leading to increased cell growth or replication. She also assessed the impact of vibrations on cell proliferation by culturing vascular SMCs in growth environments simulating disturbed flow, with high-frequency vibrations and small amplitude motions. Among different combination of the two parameters, she found that only cells exposed to a displacement of 12 μm at 45 Hz exhibited significantly higher growth rates compared to cells cultured in static conditions. This suggests that both the frequency and amplitude of the stimulus may impact on cellular functions. At variance to this investigation, a study conducted

in 2012[64] demonstrated that two murine myoblast cell lines, C2C12 and L6C11, exhibited reduced proliferation rates compared to control cells after being exposed to 30 Hz-frequency vibrations for one week. The cell index, a measure of cell quantity in a culture plate well, was significantly lower in treated wells. They also concluded that the decreased growth rate was not due to apoptosis activation, since no differences in apoptotic cell percentages were observed. Therefore, they suggested that mechanical vibration might induce micro cytoskeletal alterations, leading to a reduction in cycling capabilities of cells. This discrepancy among the studies available in the literature need further investigation to understand more in detail the effect of high-frequency vibrations in this cell phenotype.

High-frequency mechanical stimuli modulate gene expression in vascular cells

Exposure to high-frequency stimuli may modulate the expression level of some genes in vascular cells. It is known that altered blood flow hemodynamics triggers an adaptive response of the vessel wall involving migration and proliferation of vascular SMCs in the subintimal space[65] through the activation of intracellular signaling molecules such as MAPKs.[66,67] The MAPK cascade, comprising ERK1 and ERK2, is well known for its role in mediating signal transduction induced by hemodynamic forces and growth factors.[68] This cascade activates the nuclear transcription factor Elk-1, crucial for the transcriptional activation of early growth response 1 (Egr-1),[69] which coordinates the expression of various endothelial and vascular smooth muscle cell proteins involved in the molecular signaling activated by hemodynamic cues, such as PDGF-A, PDGF-B, and transforming growth factor-β (TGF-b).[68,70] Based on this rationale, Loth et al.[68] examined the distribution and activity level of ERK1/2 in a porcine arteriovenous graft model. This study showed that venous anastomosis exhibits areas of high-frequency vein-wall vibration (~300 Hz), which correlate with elevated activity levels of ERK1/2 and intimal thickening. In detail, a densitometric analysis revealed that ERK1/2 primarily localized in regions with the highest intensity of vibration within the intimal and medial SMCs. Molecular investigations using MAPK assay and Western Blot assay confirmed their enhanced activity along with increased activation of the downstream effector Elk-1. These findings provide initial evidence of a potential association between vein-wall vibration and the ERK1/2 mechanotransduction pathway. Furthermore, 4 weeks post-anastomosis, there was a modest increase in T lymphocyte and macrophage infiltration across the anastomotic regions, albeit not co-localized with activated ERK1/2 within the intima.[68] This suggests the possible existence of an ERK1/2-independent mechanism responsible for recruiting inflammatory cells to the damaged vessel site, although this aspect was not investigated in this study. Overall, these findings suggest that mechanical variables, such as turbulence-induced vein-wall vibrations, may significantly influence MAPK activation in intimal endothelial cells. However, given the proximity of the oscillation region to the flow branching area, it is challenging to distinguish their individual effects. Therefore, further research is necessary to elucidate the precise distribution of ERK1/2 and its role in mechanotransduction.

Another signaling molecule that has a pivotal role in regulating blood flow and oxygenation of tissues is nitric oxide (NO). Nitric oxide is produced and released into the blood by the endothelial cells that line the blood vessels and lymphatic vessels.[71] The effect of high-frequencies stimuli on NO release by ECs was already investigated in the early 1990s by Hutchenson,[72] who found that the molecule is produced as a function of pulsatility with an optimal frequency of pulsation of 2–8 Hz (120–480 cpm). The baseline pulsations in the human circulation are in the frequency range of 1–2 Hz and additional pulsations beyond these increase NO bioavailability via eNOS.[72–74] Vibration stimulates ECs to produce and release NO and crucial for its generation is the endothelial nitric oxide synthase. Despite the precise mechanisms by which vibrations influence endothelial mechanosensors to modulate eNOS activity are not fully understood, it seems that the endothelial cells mechanosensor-proteins Syndecan-4 (Syn4), VEGF, and KLF2 translate the physical force from the vibration into biochemical signals. Studies have demonstrated that vibroacoustic stimulation at 100 Hz induces Syn4 and VEGF expression.[75,76] NO, in response, regulates blood flow and vascular tone by affecting vascular smooth muscle cells, activating guanylate cyclase (sGC),[77] and regulates the phosphorylation of ERK1/2.[78] Significant NO-release increment has been observed with different types of vibrations, including whole-body periodic acceleration at 6 Hz,[79] arm-applied vibration at 50 Hz,[80] chest-applied sonic vibration at 100 Hz,[81] and low amplitude-vibrations at various body surfaces ranging from 150 to 250 Hz.[82] Furthermore, nuclei of endothelial, smooth muscle, and adventitial cells from rat ventral tail arteries exposed for 4 h to a 60 Hz-vibration, showed an increased immunostaining of the $Ca+^2$ activated nuclear factor of activated T-cells cytoplasmic 3 (NFATc3), induced by the upregulation and nuclear translocation of the factor.[47] Since NFAT is known to regulate cardiac hypertrophy,[83] it is plausible to consider its involvement in vibration-induced endothelial cell injury. Although the mechanism of action and protein expression influenced by NFAT were not characterized, evidence concerning this factor

could be relevant for future research aimed at investigating the cellular and molecular mechanisms involved in the early stages of vibration injury.

Recent research suggests that high-frequency mechanical vibration can promote inflammation and oxidative stress. Indeed, the stimulation of cultured cells increases the expression of the inflammatory factor NF-κB,[57] and the inflammatory cytokines tumour necrosis factor alpha (TNF-α), interleukin-8 (IL-8), interleukin-4 (IL-4), and interleukin-10 (IL-10),[84] whereas the ventral tail artery of rats exposed to vibrations show a higher expression of interleukin-6 (IL-6), interleukin-1β (IL-1β), and metallothionein 1a (MT-1a).[63]

In addition, vibrations at 30 Hz have an effect on the muscle atrophy pathway in myoblasts, which exhibit a reduced expression of myostatin, a key gene involved in muscular tissue loss.[64,85–89] In vivo, a decreased expression of both myostatin and atrogin-1, the other master gene of the atrophy pathway, was observed after 30 Hz-vibration exposure, and a significant phosphorylation of protein kinase B (AKT) was found.[64] Activation of the Akt/mTOR pathway has been shown as necessary and sufficient to induce hypertrophy and to block skeletal muscle atrophy.[85,89,90] Therefore, high-frequency vibrations appear to induce hypertrophy in skeletal muscle.[64] The same study revealed an enhanced expression of M-cadherin,[64] which has been shown to be crucial in regulating myoblast alignment and fusion.[91]

Furthermore, in 2018, Krajnak et al.[92] found a rise in the transcript number for the genes metallothionen 1a (*mt1a*), intracellular adhesion molecule-1 (*i-cam1*), and myeloid leukemia-1 protein (*runx*) by analysing tissue samples from the ventral tail artery of rats subjected for several days to a 250 Hz-vibration.

A more recent work[84] investigated the effects of vibration on the expression of long non-coding RNA (IncRNA) maternally expressed gene 3 (MEG3) by vascular endothelial cells in vitro. lncRNA MEG3 is involved in the physiological and pathological processes of various vascular diseases and could be considered an effective indicator of vascular structural and functional changes. A vibration-induced decrease in lncRNA MEG3 expression was observed after 1-day stimulation with three different frequencies (63, 200, and 250 Hz), while in the 2-day exposure group, this decrease was evident only with the 63 Hz vibration. Based on these findings, different frequencies and durations of the stimulus seem to differently affect cell expression. However, to date, the association between vibration-induced endothelial cell damage and lncRNA MEG3 remains unclear; therefore, further exploration of its involvement in the process is needed (Table 1).

Conclusions

In light of all the in vivo and in vitro studies here reported, there is evidence that mechanical vibrations affect cellular phenotypes and gene expression of vascular cells. Specifically, high-frequency vibrations have been shown to induce morphological and functional changes in vascular cells, including alterations in cytoskeletal organization, proliferation, and production of signaling factors. These alterations may contribute to vascular impairment, causing vascular constriction and disruption of the internal elastic lamina, as well as the induction of inflammatory responses, which together might contribute to vascular remodeling and occlusion. Key effectors and signaling pathways, such as ERK1/2, that in SMCs lead to intimal thickening and recruitment of inflammatory cells to damaged vessel sites, Syn4, VEGF, KLF2, ICAM-1, and NFATc3, have been identified in responding to high-frequency vibrations. Furthermore, this mechanical stimulus has been found to influence muscle atrophy pathways, potentially leading to hypertrophic growth of skeletal muscle which could explain stenosis development occurring in some vascular diseases. Whether similar effects develop also in vascular vessel wall subjected to these mechanical stimuli needs further investigation.

It is well-established that disturbances in vascular stresses and strains, whether arising from physiological activities or pathological conditions, can impact the mechanosensors and signaling pathways of endothelial and smooth muscle cells. This review underscores that cellular responses to these cues exhibit variability contingent upon factors like the frequency, duration, and intensity of the stimulus, highlighting the complexity of mechanotransduction processes in vascular cells.

Despite the wealth of evidence suggesting the possible role of high-frequency vibration in vascular damage, several gaps in understanding remain, particularly concerning the molecular mechanisms underlying cellular responses to this stimulus and its implications for vessels disease. Therefore, further research is warranted to elucidate these mechanisms of mechanotransduction and their potential therapeutic implications in vascular pathologies. For instance, studying the effect of in vitro exposure of vascular cells to vibrations at the frequencies observed through computational studies could be useful for the comprehension of the cellular effects triggered by this type of stimulus at molecular and protein level. In addition, also in vivo observation of blood vessels subjected to vibrations could allow the investigation of the contribution of different vascular cells to the biological response. Finally, understanding more in detail the mechanism by which high-frequency vibrations induce vascular changes may open the possibility of developing new pharmacological interventions for vascular diseases.

Table 1. Main effects of high-frequency stimuli on vascular cells.

Model	Method of stimulus application	Stimulus	Amplitude or acceleration	Duration of the stimulus	Observed characteristics	References
Morphological changes						
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating platform	125 Hz vibration	49 m/s^2	4 h	No signs of intimal and medial trauma; large involution in IEL; ECs compression	Krajnak et al.[46]
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating stage	60 Hz vibration	49 m/s^2	9 days, 4 h/day	Loss and thinning of ECs; activated platelets coating the exposed subendothelial tissue; vacuoles formation in ECs and SMCs; IEL disruption	Curry et al.[47]
Rat embryonic and human dermal fibroblasts	Cell cultured on an elastic membrane horizontally stretched	0.0001–20 Hz cyclic strain	1%–15%	~8 h	Cell orientation	Jungbauer et al.[53]
ECs	Cells cultured on a silicon membrane mounted on a vertical shaker	31.5 Hz cyclic strain	19, 36, 50, and 63 m/s^2	5 min	Changes in cell morphology and decreased distribution density	Mu et al.[57]
Functional changes						
Rat hind leg artery	Rat's hind leg strapped to a vertically oscillating platform	30 and 480 Hz vibration	5 g	30 days, 4 h/day	Cell proliferation and intimal thickening; IEL disruption	Inaba et al.[60]
Rat hind leg artery	Rat's hind leg strapped to a vertically oscillating platform	60 Hz vibration	5 g	90 days	Cell proliferation and intimal thickening; IEL disruption	Okada et al.[62]
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating platform	62.5, 125, and 250 Hz vibration	49 m/s^2	10 days, 4 h/days	Vascular SM thickening after 250 Hz stimulation	Krajnak et al.[63]
Rat aortic SMCs	Cell culture plate mounted on a vertical shaker	15, 30, and 45 Hz vibration	0.004, 0.008, 0.012 mm	4 h	↑ Proliferation after 45 Hz-frequency 0.012 mm-amplitude stimulation	Bittle[59]
Murine myoblast cell lines	Cell culture plate mounted on a vertical shaker	30 Hz vibration	11 mm	7 days, 1 h/day	↓ Proliferation	Ceccarelli et al.[64]

(Continued)

Table 1. (Continued)

Model	Method of stimulus application	Stimulus	Amplitude or acceleration	Duration of the stimulus	Observed characteristics	References
Gene expression						
Porcine AV graft	Flow-induced vibrations of the vein-wall after porcine AV graft-vein anastomosis creation	~300 Hz vibration	NR	4 h and 4 weeks	↑ERK1/2	Loth et al.[68]
Human coronary ECs	Vibroacustic stimulation (VATS)	100 Hz vibration	NR	2 days, 2 times/day, 30 min/time	↑Syn4, VEGF, KLF-2, eNOS, and p-eNOS	Uryash and Adams[75]
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating stage	60 Hz vibration	49 m/s^2	45 min	NFATc3	Curry et al.[47]
ECs	Cells cultured on a silicon membrane mounted on a vertical shaker	31.5 Hz cyclic strain	19, 36, 50, and 63 m/s^2	5 min	↑NF-kB	Mu et al.[57]
Human umbilical vein ECs	Cell culture plate mounted on a vibration platform	63, 200, 250 Hz vibration	6.76, 5.08, and 4.56 m/s^2	1/2 days, 1–4 h/day	↑ TNF-α, IL-8, IL-4, IL-10, ↓ MEG3 after 1-day stimulation, and ↓ MEG3 after 2-day stimulation at 63 Hz	Hongyu et al.[84]
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating platform	62.5, 125, and 250 Hz vibration	49 m/s^2	10 days, 4 h/days	↑ IL-6, MT-1a, and ↑ IL-1β after stimulation at 62.5 and 125 Hz	Krajnak et al.[63]
Murine myoblast cell lines	Cell culture plate mounted on a vertical shaker	30 Hz vibration	11 mm	7 days, 1 h/day	↓ Myostatin and ↑ M-cadherin	Ceccarelli et al.[64]
Murine tybialis anterior muscle	Rat's cage mounted on a vertical shaker	30 Hz vibration	11 mm	3 weeks, 1 h/day	↓ Myostatin, Atrogin-1, and ↑ P-Akt	Ceccarelli et al.[64]
Rat ventral tail artery	Rat's tails strapped to a vertically oscillating platform	62.5 and 250 Hz vibration	49 m/s^2	10 days, 4 h/day	↑ *mt1a*, *icam-1*, and *runx*	Krajnak et al.[92]

Symbol ↑: upregulation/activation; ↓: downregulation/inhibition; NR: not reported.

Declaration of conflicting interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) received no financial support for the research, authorship, and/or publication of this article.

ORCID iDs

Elena Carrara https://orcid.org/0009-0008-8513-5962
Luca Soliveri https://orcid.org/0000-0002-8096-2925
Michela Bozzetto https://orcid.org/0000-0002-2045-5550

References

1. Li YSJ, Haga JH and Chien S. Molecular basis of the effects of shear stress on vascular endothelial cells. *J Biomech* 2005; 38(10): 1949–1971.
2. Baeyens N, Bandyopadhyay C, Coon BG, et al. Endothelial fluid shear stress sensing in vascular health and disease. *J Clin Invest* 2016; 126(3): 821–828.
3. Humphrey JD and Schwartz MA. Vascular mechanobiology: homeostasis, adaptation, and disease. *Annu Rev Biomed Eng* 2021; 23(1): 1–27.
4. Chien S. Mechanotransduction and endothelial cell homeostasis: the wisdom of the cell. *Am J Physiol-Heart Circ Physiol* 2007; 292(3): H1209–H1224.
5. Brahmbhatt A, Remuzzi A, Franzoni M, et al. The molecular mechanisms of hemodialysis vascular access failure. *Kidney Int* 2016; 89(2): 303–316.
6. Franzoni M, Cattaneo I, Longaretti L, et al. Endothelial cell activation by hemodynamic shear stress derived from arteriovenous fistula for hemodialysis access. *Am J Physiol Heart Circ Physiol* 2016; 310(1): H49–H59.
7. Cao W, Zhang D, Li Q, et al. Biomechanical stretch induces inflammation, proliferation, and migration by activating NFAT5 in arterial smooth muscle cells. *Inflammation* 2017; 40(6): 2129–2136.
8. Wang Y, Cao W, Cui J, et al. Arterial wall stress induces phenotypic switching of arterial smooth muscle cells in vascular remodeling by activating the YAP/TAZ signaling pathway. *Cell Physiol Biochem* 2018; 51(2): 842–853.
9. Chiu JJ and Chien S. Effects of disturbed flow on vascular endothelium: pathophysiological basis and clinical perspectives. *Physiol Rev* 2011; 91(1): 327–387.
10. Souche A and Valen-Sendstad K. High-fidelity fluid structure interaction simulations of turbulent-like aneurysm flows reveals high-frequency narrowband wall vibrations: a stimulus of mechanobiological relevance? *J Biomech* 2022; 145: 111369.
11. Khan MO, Toro Arana V, Najafi M, et al. On the prevalence of flow instabilities from high-fidelity computational fluid dynamics of intracranial bifurcation aneurysms. *J Biomech* 2021; 127: 110683.
12. Valen-Sendstad K, Piccinelli M and Steinman DA. High-resolution computational fluid dynamics detects flow instabilities in the carotid siphon: implications for aneurysm initiation and rupture? *J Biomech* 2014; 47(12): 3210–3216.
13. Bozzetto M, Ene-Iordache B and Remuzzi A. Transitional flow in the venous side of patient-specific arteriovenous fistulae for hemodialysis. *Ann Biomed Eng* 2016; 44(8): 2388–2401.
14. Soliveri L, Bozzetto M, Brambilla P, et al. Hemodynamics in AVF over time: a protective role of vascular remodeling toward flow stabilization. *Int J Artif Organs* 2023; 46(10–11): 547–554.
15. Roy-Chaudhury P, Arend L, Zhang J, et al. Neointimal hyperplasia in early arteriovenous fistula failure. *Am J Kidney Dis* 2007; 50(5): 782–790.
16. Viecelli AK, Mori TA, Roy-Chaudhury P, et al. The pathogenesis of hemodialysis vascular access failure and systemic therapies for its prevention: optimism unfulfilled. *Semin Dial* 2018; 31(3): 244–257.
17. Sabiu G and Gallieni M. Pathophysiology of arteriovenous fistula maturation and nonmaturation. *Clin J Am Soc Nephrol* 2023; 18(1): 8.
18. Bozzetto M, Remuzzi A and Valen-Sendstad K. Flow-induced high frequency vascular wall vibrations in an arteriovenous fistula: a specific stimulus for stenosis development? *Phys Eng Sci Med* 2024; 47(1): 187–197.
19. Bruneau DA, Valen-Sendstad K and Steinman DA. Onset and nature of flow-induced vibrations in cerebral aneurysms via fluid-structure interaction simulations. *Biomech Model Mechanobiol* 2023; 22(3): 761–771.
20. Bruneau DA, Steinman DA and Valen-Sendstad K. Understanding intracranial aneurysm sounds via high-fidelity fluid-structure-interaction modelling. *Commun Med* 2023; 3(1): 163.
21. Ferguson GG. Turbulence in human intracranial saccular aneurysms. *J Neurosurg* 1970; 33(5): 485–497.
22. Aaslid R and Nornes H. Musical murmurs in human cerebral arteries after subarachnoid hemorrhage. *J Neurosurg* 1984; 60(1): 32–36.
23. Roach MR. An experimental study of the production and time course of poststenotic dilatation in the femoral and carotid arteries of adult dogs. *Circ Res* 1963; 13(6): 537–551.
24. Wang JZ, Tie B, Welkowitz W, et al. Modeling sound generation in stenosed coronary arteries. *IEEE Trans Biomed Eng* 1990; 37(11): 1087–1094.
25. Foreman JEK and Hutchison KJ. Arterial wall vibration distal to stenoses in isolated arteries of dog and man. *Circ Res* 1970; 26(5): 583–590.
26. Simkins TE and Stehbens WE. Vibrations recorded from the adventitial surface of experimental aneurysms and arteriovenous fistulas. *Vasc Surg* 1974; 8(3): 153–165.
27. Fillinger MF, Reinitz ER, Schwartz RA, et al. Graft geometry and venous intimal-medial hyperplasia in arteriovenous loop grafts. *J Vasc Surg* 1990; 11(4): 556–566.
28. Seo JH and Mittal R. A coupled flow-acoustic computational study of bruits from a modeled stenosed artery. *Med Biol Eng Comput* 2012; 50(10): 1025–1035.
29. Ozden K, Sert C and Yazicioglu Y. Effect of stenosis shape on the sound emitted from a constricted blood vessel. *Med Biol Eng Comput* 2020; 58(3): 643–658.
30. Mancini V, Tommasin D, Li Y, et al. Detecting carotid stenosis from skin vibrations using laser doppler vibrometry – an in vitro proof-of-concept. *PLoS One* 2019; 14(6): e0218317.

31. Balasso A, Fritzsche M, Liepsch D, et al. High-frequency wall vibrations in a cerebral patient-specific aneurysm model. *Biomed Eng Biomed Tech* 2019; 64(3): 275–284.
32. Ozturk N, Erkan O, Uslu S, et al. Role of adenosine triphosphate and protein kinase A in the force-frequency relationship in isolated rat cardiomyocytes. *Arch Biol Sci* 2023; 75(1): 47–56.
33. Burridge K, Monaghan-Benson E and Graham DM. Mechanotransduction: from the cell surface to the nucleus via RhoA. *Philos Trans R Soc B Biol Sci* 2019; 374(1779): 20180229.
34. Givens C and Tzima E. Endothelial mechanosignaling: does one sensor fit all? *Antioxid Redox Signal* 2016; 25(7): 373–388.
35. Poelmann RE and Gittenberger-de Groot AC. Hemodynamics in cardiac development. *J Cardiovasc Dev Dis* 2018; 5(4): 54.
36. Nilius B, Viana F and Droogmans G. Ion channels in vascular endothelium. *Annu Rev Physiol* 1997; 59: 145–170.
37. Olesen SP, Claphamt D and Davies P. Haemodynamic shear stress activates a K+ current in vascular endothelial cells. *Nature* 1988; 331(6152): 168–170.
38. Schwarz G, Droogmans G and Nilius B. Shear stress induced membrane currents and calcium transients in human vascular endothelial cells. *Pflugers Arch* 1992; 421(4): 394–396.
39. Baeyens N, Mulligan-Kehoe MJ, Corti F, et al. Syndecan 4 is required for endothelial alignment in flow and atheroprotective signaling. *Proc Natl Acad Sci* 2014; 111(48): 17308–17313.
40. Chen J, Green J, Yurdagul A, et al. αvβ3 integrins mediate flow-induced NF-κB activation, proinflammatory gene expression, and early atherogenic inflammation. *Am J Pathol* 2015; 185(9): 2575–2589.
41. Tzima E, Irani-Tehrani M, Kiosses WB, et al. A mechanosensory complex that mediates the endothelial cell response to fluid shear stress. *Nature* 2005; 437(7057): 426–431.
42. Tzima E, del Pozo MA, Shattil SJ, et al. Activation of integrins in endothelial cells by fluid shear stress mediates Rho-dependent cytoskeletal alignment. *EMBO J* 2001; 20(17): 4639–4647.
43. Yamamoto K and Ando J. New molecular mechanisms for cardiovascular disease: blood flow sensing mechanism in vascular endothelial cells. *J Pharmacol Sci* 2011; 116(4): 323–331.
44. Jetta D, Gottlieb PA, Verma D, et al. Shear stress-induced nuclear shrinkage through activation of Piezo1 channels in epithelial cells. *J Cell Sci* 2019; 132(11): jcs226076.
45. Noguchi T, Matozaki T, Horita K, et al. Role of SH-PTP2, a protein-tyrosine phosphatase with Src homology 2 domains, in insulin-stimulated Ras activation. *Mol Cell Biol* 1994; 14(10): 6674–6682.
46. Krajnak K, Dong RG, Flavahan S, et al. Acute vibration increases α_{2C} -adrenergic smooth muscle constriction and alters thermosensitivity of cutaneous arteries. *J Appl Physiol* 2006; 100(4): 1230–1237.
47. Curry BD, Bain JLW, Yan J, et al. Vibration injury damages arterial endothelial cells. *Muscle Nerve* 2002; 25(4): 527–534.
48. Joris I and Majno G. Medial changes in arterial spasm induced by L-norepinephrine. *Am J Pathol* 1981; 105(3): 212–222.
49. Rabinovitch M. Cell-extracellular matrix interactions in the ductus arteriosus and perinatal pulmonary circulation. *Semin Perinatol* 1996; 20(6): 531–541.
50. Liu B, Qu MJ, Qin KR, et al. Role of cyclic strain frequency in regulating the alignment of vascular smooth muscle cells in vitro. *Biophys J* 2008; 94(4): 1497–1507.
51. Kanda K, Matsuda T and Oka T. Two-dimensional orientational response of smooth muscle cells to cyclic stretching. *ASAIO J* 1992; 38(3): M382.
52. Hsu HJ, Lee CF and Kaunas R. A dynamic stochastic model of frequency-dependent stress fiber alignment induced by cyclic stretch. *PLoS One* 2009; 4(3): e4853.
53. Jungbauer S, Gao H, Spatz JP, et al. Two characteristic regimes in frequency-dependent dynamic reorientation of fibroblasts on cyclically stretched substrates. *Biophys J* 2008; 95(7): 3470–3478.
54. Finlay HM, Whittaker P and Canham PB. Collagen organization in the branching region of human brain arteries. *Stroke* 1998; 29(8): 1595–1601.
55. Standley PR, Camaratta A, Nolan BP, et al. Cyclic stretch induces vascular smooth muscle cell alignment via NO signaling. *Am J Physiol-Heart Circ Physiol* 2002; 283(5): H1907–H1914.
56. Greiner AM, Biela SA, Chen H, et al. Featured article: temporal responses of human endothelial and smooth muscle cells exposed to uniaxial cyclic tensile strain. *Exp Biol Med* 2015; 240(10): 1298–1309.
57. Mu L, Sun A, Chen Y, et al. Vascular response to the microcirculation in the fingertip by local vibration with varied amplitude. *Front Bioeng Biotechnol* 2023; 11: 1197772.
58. Ingber D. Mechanobiology and diseases of mechanotransduction. *Ann Med* 2003; 35(8): 564–577.
59. Bittle B. An investigation into the role of arterial wall vibration in the pathogenesis of atherosclerosis, https://dr.lib.iastate.edu/handle/20.500.12876/63696 (1994, accessed 5 April 2024).
60. Inaba R, Furuno T and Okada A. Effects of low- and high-frequency local vibration on the occurrence of intimal thickening of the peripheral arteries of rats. *Scand J Work Environ Health* 1988; 14(5): 312–316.
61. Nilsson T, Wahlström J and Burström L. Hand-arm vibration and the risk of vascular and neurological diseases—a systematic review and meta-analysis. *PLoS One* 2017; 12(7): e0180795.
62. Okada A, Inaba R, Furuno T, et al. Usefulness of blood parameters, especially viscosity, for the diagnosis and elucidation of pathogenic mechanisms of the hand-arm vibration syndrome. *Scand J Work Environ Health* 1987; 13(4): 358–362.
63. Krajnak K, Miller GR, Waugh S, et al. Characterization of frequency-dependent responses of the vascular system to repetitive vibration. *J Occup Environ Med*. 2010; 52(6): 584–594.
64. Ceccarelli G, Benedetti L, Galli D, et al. Low-amplitude high frequency vibration down-regulates myostatin and atrogin-1 expression, two components of the atrophy pathway in muscle cells: down-regulation of myostatin and atrogin-1 expression. *J Tissue Eng Regen Med*. 2014; 8(5): 396–406.
65. Glagov S. Intimal hyperplasia, vascular modeling, and the restenosis problem. *Circulation* 1994; 89(6): 2888–2891.

66. Zou Y, Hu Y, Metzler B, et al. Signal transduction in arteriosclerosis: mechanical stress-activated MAP kinases in vascular smooth muscle cells (review). *Int J Mol Med* 1998; 1(5): 827–861.
67. Traub O and Berk BC. Laminar shear stress. *Arterioscler Thromb Vasc Biol* 1998; 18(5): 677–685.
68. Loth F, Fischer PF, Arslan N, et al. Transitional flow at the venous anastomosis of an arteriovenous graft: potential activation of the ERK1/2 mechanotransduction pathway. *J Biomech Eng* 2003; 125(1): 49–61.
69. Schwachtgen JL, Houston P, Campbell C, et al. Fluid shear stress activation of egr-1 transcription in cultured human endothelial and epithelial cells is mediated via the extracellular signal-related kinase 1/2 mitogen-activated protein kinase pathway. *J Clin Invest* 1998; 101(11): 2540–2549.
70. Song RH, Kocharyan HK, Fortunato JE, et al. Increased flow and shear stress enhance in vivo transforming growth factor-β1 after experimental arterial injury. *Arterioscler Thromb Vasc Biol* 2000; 20(4): 923–930.
71. Bartel L and Mosabbir A. Possible mechanisms for the effects of sound vibration on human health. *Healthcare* 2021; 9(5): 597.
72. Hutcheson IR and Griffith TM. Release of endothelium-derived relaxing factor is modulated both by frequency and amplitude of pulsatile flow. *Am J Physiol-Heart Circ Physiol* 1991; 261(1): H257–H262.
73. Stefano GB, Prevot V, Cadet P, et al. Vascular pulsations stimulating nitric oxide release during cyclic exercise may benefit health: a molecular approach (review). *Int J Mol Med* 2001; 7(2): 119–129.
74. Adams JA, Uryash A and Lopez JR. Non-invasive pulsatile shear stress modifies endothelial activation; a narrative review. *Biomedicines* 2022; 10(12): 3050.
75. Uryash A and Adams JA. Abstract 18011: vibroacoustic non-invasive stimulation (VATS) of human coronary endothelial cells induced syndecan-4, VEGF and KLF2 mechanosensor control of eNOS. *Circulation* 2017; 136(suppl_1): A18011.
76. Uryash A and Adams JA. Abstract 17906: wearable vibroacoustic transthoracic stimulation (VATS) provides cardioprotection by syndecan-4 mechanosensor regulation of NFAT, JNK/ERK in rats after myocardial infarction. *Circulation* 2017; 136(suppl_1): A17906.
77. Chen K, Pittman RN and Popel AS. Nitric oxide in the vasculature: where does it come from and where does it go? A quantitative perspective. *Antioxid Redox Signal* 2008; 10(7): 1185–1198.
78. White CR, Haidekker MA, Stevens HY, et al. Extracellular signal-regulated kinase activation and endothelin-1 production in human endothelial cells exposed to vibration. *J Physiol* 2004; 555(2): 565–572.
79. Uryash A, Wu H, Bassuk J, et al. Low-amplitude pulses to the circulation through periodic acceleration induces endothelial-dependent vasodilatation. *J Appl Physiol* 2009; 106(6): 1840–1847.
80. Maloney-Hinds C, Petrofsky JS, Zimmerman G, et al. The role of nitric oxide in skin blood flow increases due to vibration in healthy adults and adults with type 2 diabetes. *Diabetes Technol Ther* 2009; 11(1): 39–43.
81. Uryash A and Adams JA. Abstract 17052: wearable vibroacoustic transthoracic stimulation improves left ventricular function, remodeling and regulates syndecan-4/VEGF levels in rats after myocardial infarction. *Circulation* 2016; 134(suppl_1): A17052.
82. Skoglund CR. Vasodilatation in human skin induced by low-amplitude high-frequency vibration. *Clin Physiol* 1989; 9(4): 361–372.
83. Musarò A, McCullagh KJA, Naya FJ, et al. IGF-1 induces skeletal myocyte hypertrophy through calcineurin in association with GATA-2 and NF-ATc1. *Nature* 1999; 400(6744): 581–585.
84. Hongyu Y, Qingsong C, Zheng LI, et al. Effects of vibration on expressions of vascular endothelial inflammatory factors and lncRNA *MEG3 in vitro*. *J Environ Occup Med* 2022; 39(11): 1209–1213.
85. Bodine SC, Stitt TN, Gonzalez M, et al. Akt/mTOR pathway is a crucial regulator of skeletal muscle hypertrophy and can prevent muscle atrophy in vivo. *Nat Cell Biol* 2001; 3(11): 1014–1019.
86. Lee SJ and McPherron AC. Regulation of myostatin activity and muscle growth. *Proc Natl Acad Sci* 2001; 98(16): 9306–9311.
87. Gomes MD, Lecker SH, Jagoe RT, et al. Atrogin-1, a muscle-specific F-box protein highly expressed during muscle atrophy. *Proc Natl Acad Sci* 2001; 98(25): 14440–14445.
88. Kandarian SC and Jackman RW. Intracellular signaling during skeletal muscle atrophy. *Muscle Nerve* 2006; 33(2): 155–165.
89. Sandri M. Signaling in muscle atrophy and hypertrophy. *Physiology* 2008; 23(3): 160–170.
90. Bodine SC, Latres E, Baumhueter S, et al. Identification of ubiquitin ligases required for skeletal muscle atrophy. *Science* 2001; 294(5547): 1704–1708.
91. Wang Y, Mohamed JS and Alway SE. M-cadherin-inhibited phosphorylation of ß-catenin augments differentiation of mouse myoblasts. *Cell Tissue Res* 2013; 351(1): 183–200.
92. Krajnak K, Miller GR and Waugh S. Contact area affects frequency-dependent responses to vibration in the peripheral vascular and sensorineural systems. *J Toxicol Environ Health A* 2018; 81(1–3): 6–19.