JOURNAL OF THE AMERICAN COLLEGE OF CARDIOLOGY VOL. 75, NO. 8, 2020

JACC FOCUS SEMINAR: VASCULAR AGING

Biological Versus Chronological Aging

JACC Focus Seminar

Magda R. Hamczyk, PhD,[a,b,c] Rosa M. Nevado, MS,[a,b] Ana Barettino, MS,[a,b] Valentín Fuster, MD, PhD,[a,d] Vicente Andrés, PhD[a,b]

ABSTRACT

Aging is the main risk factor for vascular disease and ensuing cardiovascular and cerebrovascular events, the leading causes of death worldwide. In a progressively aging population, it is essential to develop early-life biomarkers that efficiently identify individuals who are at high risk of developing accelerated vascular damage, with the ultimate goal of improving primary prevention and reducing the health care and socioeconomic impact of age-related cardiovascular disease. Studies in experimental models and humans have identified 9 highly interconnected hallmark processes driving mammalian aging. However, strategies to extend health span and life span require understanding of interindividual differences in age-dependent functional decline, known as biological aging. This review summarizes the current knowledge on biological age biomarkers, factors influencing biological aging, and antiaging interventions, with a focus on vascular aspects of the aging process and its cardiovascular disease related manifestations.
(J Am Coll Cardiol 2020;75:919–30) © 2020 The Authors. Published by Elsevier on behalf of the American College of Cardiology Foundation. This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

Aging is the main risk factor for vascular disease and the ensuing cardiovascular and cerebrovascular events, which are the leading causes of death worldwide (1). Vascular aging entails arterial degeneration and hardening that impairs vascular function and ultimately causes end organ damage, predominantly in the heart, brain, and kidney. Age-dependent arterial injury typically manifests clinically after the fifth or sixth decade of life; however, there is a high interindividual variability in vascular disease onset and associated mortality (2). At the extremes are patients with premature aging syndromes and centenarians/supercentenarians, representing early and supernormal vascular aging, respectively (**Figure 1**) (3). The observation that individuals do not age at the same pace led to the concept of biological aging, also called functional or physiological aging. Whereas chronological aging refers only to the passage of time, biological aging relates to decline in function. In a progressively aging world population, it is critically important to define the mechanisms governing biological vascular aging in

Listen to this manuscript's audio summary by Editor-in-Chief Dr. Valentin Fuster on JACC.org.

From the [a]Centro Nacional de Investigaciones Cardiovasculares (CNIC), Spain; [b]Centro de Investigación Biomédica en Red de Enfermedades Cardiovasculares (CIBERCV), Spain; [c]Departamento de Bioquímica y Biología Molecular, Instituto Universitario de Oncología (IUOPA), Universidad de Oviedo, Oviedo, Spain; and [d]The Zena and Michael A. Wiener Cardiovascular Institute/Marie-Josée and Henry R. Kravis Center for Cardiovascular Health, Mount Sinai School of Medicine, New York, New York. The CNIC is supported by the MCNU, the ISCIII, and the Pro CNIC Foundation and is a Severo Ochoa Center of Excellence (SEV-2015-0505). Work in Dr. Andrés' laboratory is supported by the Spanish Ministerio de Ciencia, Innovación y Universidades (MCNU) (SAF2016-79490-R) and the Instituto de Salud Carlos III (ISCIII) (AC16/00091, AC17/00067, and CB16/11/00405) with cofunding from the European Regional Development Fund (ERDF/FEDER, "Una manera de hacer Europa") and the Fundació Marató TV3 (122/C/2015). Dr. Hamczyk is supported by the MCNU (post-doctoral contract FJCI-2017-33299). Ms. Nevado is supported by the Ministerio de Educación, Cultura y Deporte (pre-doctoral contract FPU16/05027). Ms. Barettino is supported by the MCNU (pre-doctoral contract BES-2017-079705). All authors have reported that they have no relationships relevant to the contents of this paper to disclose.

Manuscript received October 30, 2019; revised manuscript received November 21, 2019, accepted November 25, 2019.

ISSN 0735-1097 https://doi.org/10.1016/j.jacc.2019.11.062

ABBREVIATIONS AND ACRONYMS

AMPK = adenosine monophosphate–activated protein kinase

CACS = coronary artery calcium score

CAD = coronary artery disease

CVD = cardiovascular disease

DNAmAge = DNA methylation age

IMT = intima-media thickness

mTOR = mechanistic target of rapamycin

PWV = pulse wave velocity

T2D = type 2 diabetes

order to reduce its socioeconomic and health care burden.

BIOMARKERS OF BIOLOGICAL VASCULAR AGING

Biomarkers that truly reflect the state of vascular aging are needed to improve early detection of individuals at high risk of developing cardiovascular disease (CVD). Ideal biological age biomarkers should outperform chronological age as determinants of morbidity and mortality. Moreover, their quantification should be easy and safe, preferably in a blood test or by a noninvasive imaging technique (4). Biological age indicators described to date vary from phenotypic and functional scores to molecular biomarkers. This section discusses molecular and cellular biomarkers that reflect the decline in function of all tissues including the vasculature, functional, and structural indicators of biological vascular age and composite biomarker predictors of biological age (**Figure 2**).

MOLECULAR AND CELLULAR BIOMARKERS. Aging can be defined at the molecular and cellular level by the presence of 9 hallmarks (5), some of which, such as telomere attrition and epigenetic alterations, are commonly used to assess human biological age. Telomeres consist of repetitive DNA sequences bound by specific nucleoproteins that protect chromosome ends (6). Telomeres shorten with each cell division, and this attrition can cause cell senescence when telomeres shorten beyond a critical length (6). Age-dependent telomere shortening is associated with coronary artery disease (CAD) and predicts both all-cause and CVD-related mortality; however, it remains uncertain whether telomere attrition promotes CVD (4,6,7).

DNA methylation is a major epigenetic mechanism regulating gene expression. Various studies have identified specific CpG sites undergoing age-related changes in methylation (8,9). Hannum et al. (10) and Horvath (11) described 2 widely used DNA methylation age predictors (DNAmAges, also called epigenetic clocks) based on 71 and 353 CpGs, respectively. DNAmAge seems to be a good predictor of all-cause and CVD mortality and, to a lesser extent, incident CVD (12,13).

During aging, somatic cells accumulate mutations in their DNA, and these mutations can provide a competitive advantage, particularly to highly proliferative cells, thus leading to expansion of mutant clones and mosaicism (14). Somatic mutation-related expansion of hematopoietic cells (clonal hematopoiesis) has typically been linked to cancer; however, recent findings demonstrated an association with increased CVD risk suggesting a potential role as a major driver of atherosclerosis, thus identifying clonal hematopoiesis as a possible novel biomarker of vascular aging (14).

HIGHLIGHTS

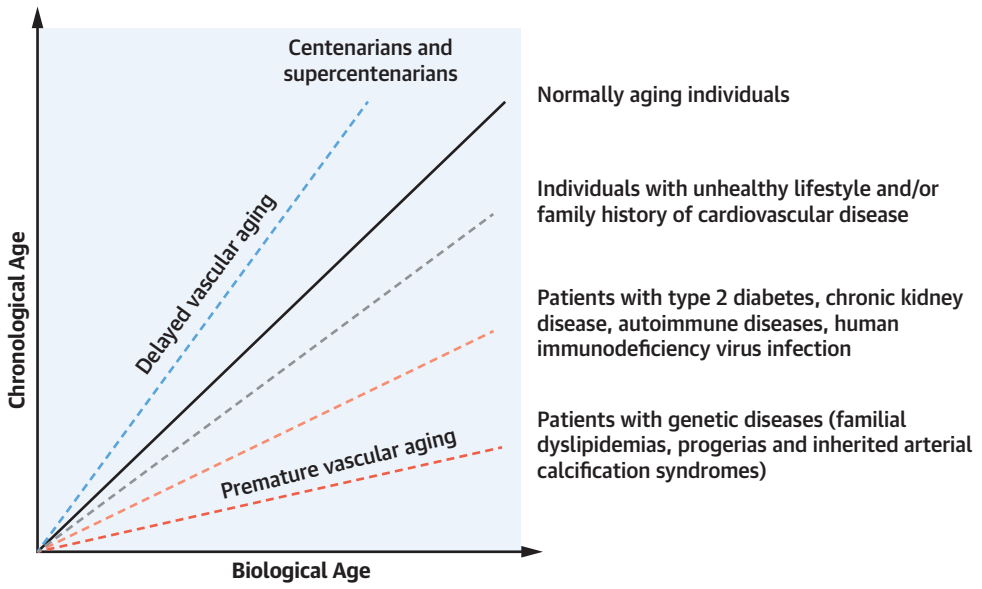
- Vascular disease onset and mortality are highly variable among individuals.
- Chronological age is suboptimal for estimating vascular aging.
- Biological vascular age should be used to select individuals for early prevention of cardiovascular disease.
- Biological vascular aging can be targeted by behavioral and pharmacological interventions.

Another characteristic of aging is chronic low-grade inflammation (inflammaging), which can be assessed by measuring circulating levels of proinflammatory molecules, such as elevated C-reactive protein and interleukin 6 (15). Inflammaging, a risk factor for many chronic diseases, including CVD, is partially driven by increased gut permeability and altered microbiota composition. Gut dysbiosis also has the potential to become a biological age estimator because it has been linked to longevity and disease (16).

Aging affects the levels of proteins, metabolites, and other biomolecules in body fluids. Higher morbidity and mortality rates have been linked to changes in individual molecules, such as insulin-like growth factor-1, growth hormone, and low-density lipoproteins (17,18). Because single molecule biomarkers tend to be oversimplistic, various high-throughput approaches have been used to analyze age-related changes in the transcriptome, proteome, and metabolome to search for a more comprehensive omics-based "blueprint" of aging, such as GlycanAge (GlycanAge Ltd., London, United Kingdom) or Metabolic Age scores (4).

VASCULAR FUNCTIONAL AND STRUCTURAL BIOMARKERS. Structural changes in aging arteries include elastin fragmentation, collagen accumulation, and medial vascular smooth muscle cell loss, causing reduced vascular compliance and increased arterial stiffness (19). The most common arterial stiffness measure is pulse wave velocity (PWV), the velocity at which the blood pressure wave moves along the arterial tree. Carotid to femoral PWV is the

FIGURE 1 Biological Versus Chronological Vascular Aging



Biological aging refers to the decline in tissue/organismal function, whereas chronological aging simply indicates the time passed since birth. In normally aging individuals, chronological age equates to biological age. Vascular aging is delayed in centenarians and supercentenarians and is accelerated in certain diseases and conditions.

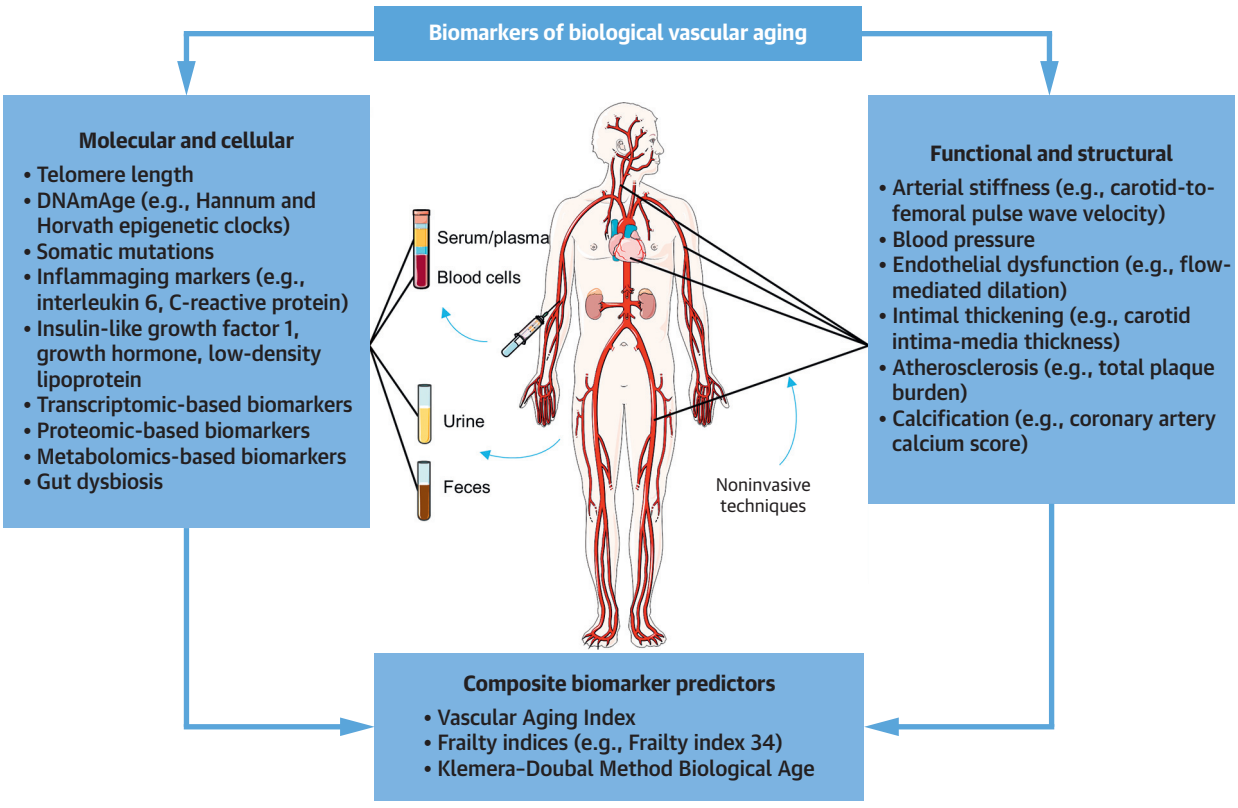
most extensively validated and standardized measure; however, brachial-ankle and heart-ankle PVW are also popular in the clinic (20,21). PWV correlates with chronological age, and a high PWV increases the risk of CVD and all-cause mortality (22,23). Arterial stiffening typically precedes and promotes hypertension, but hypertension can also accelerate arterial stiffening, indicating the existence of a positive feedback loop (24). Blood pressure also increases during aging and is associated with cardiovascular events and mortality (25).

Another common feature of vascular aging is atherosclerosis, which consists of lipid-rich plaque buildup in the intima that can lead to acute myocardial infarction or stroke. Endothelial dysfunction, a major driver of atherogenesis, can be measured by ultrasound as flow-mediated dilation (26), and flow-mediated dilation decreases during aging and is an independent predictor of CVD outcomes (26,27). Vascular ultrasound is also frequently used to measure intima-media thickness (IMT) as an estimate of subclinical atherosclerosis burden in large arteries (28,29). Carotid IMT increases with age (30) and is associated with both the prevalence and the incidence of CVD morbidity and mortality (31). However, increased IMT can also reflect nonatherosclerotic processes (30,32). More advanced atherosclerosis stages can be evaluated by quantifying various carotid plaque parameters, such as plaque presence, number, thickness, area, and volume (33), which outperform carotid IMT as a predictor of future CAD events (32).

Vascular aging is also characterized by the deposition of calcium phosphate crystals in both the arterial intima (typically related to atherosclerosis) and the media (called Mönckeberg sclerosis) (34). Both types of calcification often develop in parallel and are not always easily distinguished by imaging techniques (35). Computed tomography is the gold standard technique for quantification of coronary artery calcium score (CACS) (36). Even though CACS can be influenced by the presence of medial calcification, it is typically used as a surrogate marker for the extent of atherosclerosis because it correlates well with coronary plaque burden (37). CACS correlates with chronological age (38) and is a powerful predictor of incident CVD and all-cause mortality (39–41). An arterial age calculator based on CACS (40) is freely available online (42).

COMPOSITE BIOMARKER PREDICTORS. Age estimation based on 1 of the previously mentioned biomarkers often fails to reflect the complexity of aging and can give suboptimal assessments of biological

FIGURE 2 Biomarkers of Biological Vascular Aging



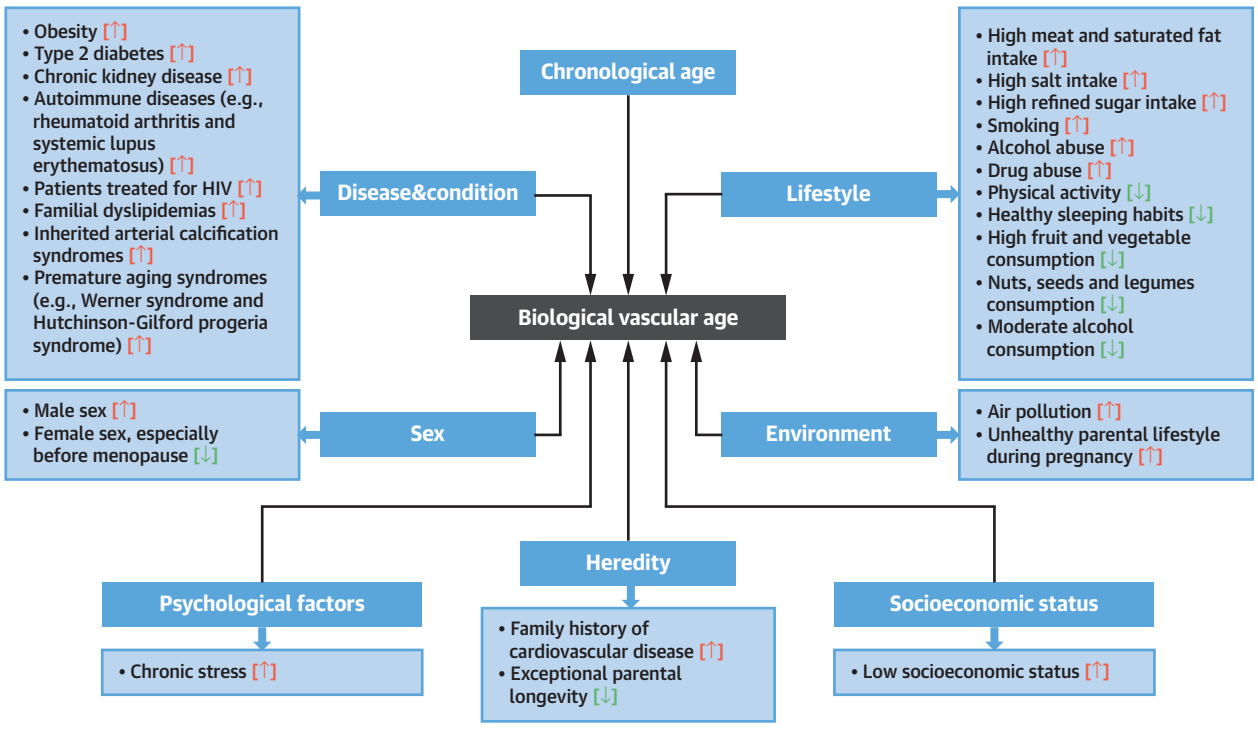
The biological age of the vasculature can be estimated using molecular and cellular biomarkers (in blood, urine, or feces), functional and structural vascular parameters (measured by noninvasive techniques), or a combination of both. This figure was created with images modified from Servier Medical Art by Servier licensed under a Creative Commons Attribution 3.0 Unsupported License. DNAmAge = DNA methylation age.

age. Therefore, better aging estimates are sought from compound scores. Composite biomarkers vary from combinations of a few biomarkers to highly complex scores developed using machine learning/artificial intelligence tools that cover multiple aspects of aging. For instance, CVD prediction is improved by the integration of carotid IMT with aortic PWV into the Vascular Aging Index (43). Likewise, Frailty Index 34, composed of 34 measures of health and function in diverse body systems, including the vasculature, outperforms DNAmAge at predicting mortality (44). Many other biological age estimators were developed by combining physical, physiological, and biochemical parameters; for example, the Klemera-Doubal Method Biological Age index is based on 10 biomarkers (45,46). However, as composite predictors of biological age become more complex, they often become impractical and costly and therefore more difficult to apply to the entire population.

FACTORS INFLUENCING BIOLOGICAL AGING OF THE VASCULAR SYSTEM

To identify factors that control biological aging, multiple studies have compared individuals with early and late onset of aging symptoms. Findings in castrated males and postmenopausal women have provided insight into the role of sex hormones in the aging process. Similarly, studies of identical twins have allowed the contribution of nonmodifiable genetic features to longevity and disease to be studied separately from the influence of environmental and lifestyle factors. Hints about longevity-promoting factors have also been obtained from research on "Blue Zones," geographic regions with an above average proportion of centenarians. Nutritional clues to healthy aging have been derived from a comparison of healthy and unhealthy diets. Additional insight into aging has come from research into conditions

FIGURE 3 Factors Influencing Biological Vascular Age



[↓] = factors decelerating biological aging; [↑] = factors accelerating biological aging; HIV = human immunodeficiency virus.

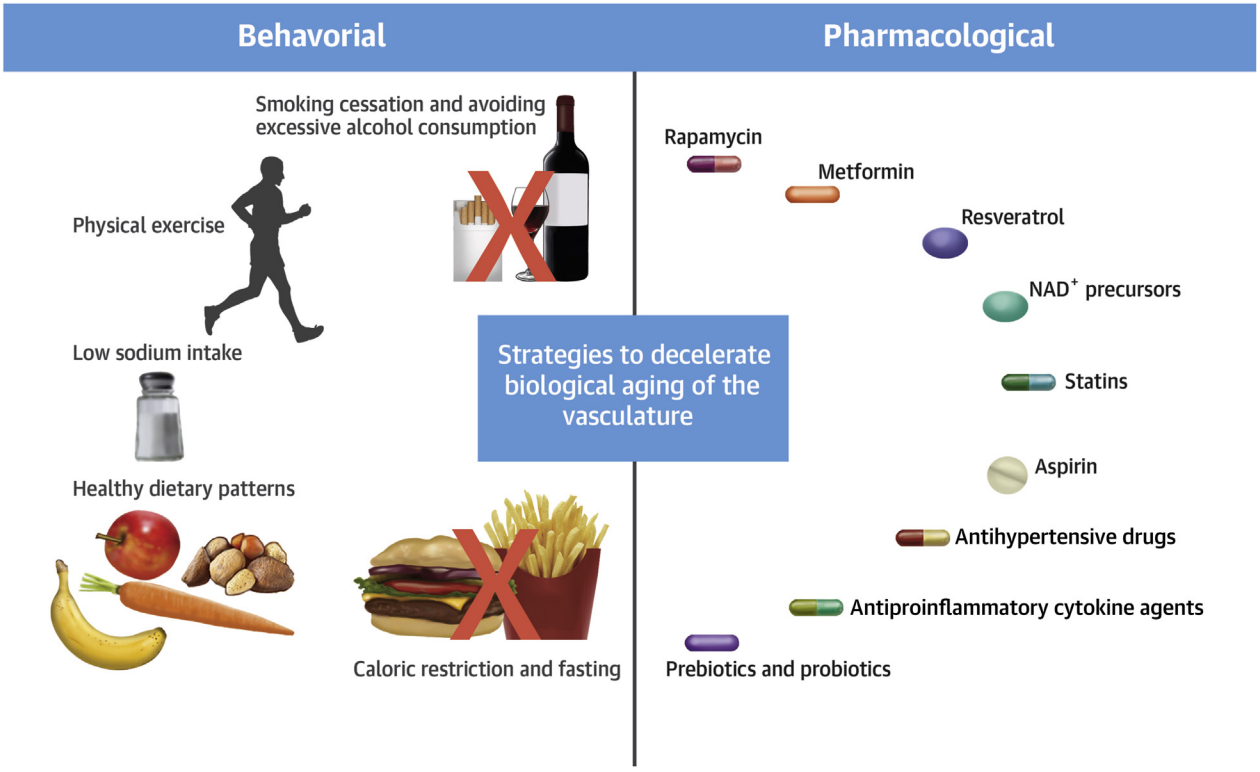
and diseases featuring a premature onset of aging. This section discusses key nonmodifiable and modifiable factors that influence vascular health and longevity as well as examples of diseases and conditions featuring early vascular impairment (**Figure 3**).

SEX AND INHERITABLE FACTORS. Women have a higher life expectancy than men, and a large majority of centenarians are women (47). Moreover, the risk of CAD is higher in men, especially below the age of 50. Accordingly, women have longer telomeres (4), lower DNAmAges (4,10,12), and lower CACSs (38) than age-matched men. This sex-dependent effect can be partially attributed to the vasculoprotective role of estrogen (47). There is also a possible adverse impact of testosterone on aging because eunuchs were reported to live longer than noncastrated men of the same socioeconomic status (48). Yet, the impact of testosterone on cardiovascular health remains controversial (49).

Health span and longevity are also crucially influenced by inheritable factors, mostly genetic and epigenetic. For example, family CVD history increases future CVD risk from 40% to 75% depending on the degree of relatedness (50). Moreover, many genetic variants are associated with an exceptionally long life span (51), and offspring of centenarians show signs of low epigenetic age in blood cells (52). There are also ethnic differences in various measures of biological age and CVD morbidity and mortality (38,53). Nevertheless, a meta-analysis of twin studies revealed that genetics accounts for only 20% to 30% of the life span variation (54), indicating that aging and longevity are significantly modulated by other factors, such as environment and lifestyle.

ENVIRONMENT AND LIFESTYLE. The influence of the environment on vascular aging begins before birth through in utero (developmental) programming (55). Parental lifestyle during pregnancy can elicit adverse effects on the long-term health of the offspring (56). Autopsy studies have detected atherosclerotic lesions in arteries of fetuses and neonates of smoker or hypercholesterolemic mothers (57,58), possibly reflecting epigenetic alterations (59). During postnatal life, the pace of biological aging can be affected by a wide range of environmental and lifestyle factors. Physical inactivity is a major mortality risk factor, and even small amounts of exercise reduce CVD and all-cause mortality (60,61). Another key modulator of vascular

CENTRAL ILLUSTRATION **Strategies to Decelerate Biological Aging of the Vascular System**



Hamczyk, M.R. et al. J Am Coll Cardiol. 2020;75(8):919-30.

See Table 1 for details. NAD^+ = nicotinamide adenine dinucleotide.

function and CVD risk is diet, with healthy aging linked to unprocessed, plant-based nutrition with moderate consumption of lean meat and fish, features of the Mediterranean and Dietary Approaches to Stop Hypertension diets (62,63). The key components of Western-type diets that promote atherosclerosis and hypertension are refined sugars, processed meat, hydrogenated vegetable oils, and high amounts of salt (62,63). Apart from the types of food consumed, the quantity of ingested food plays a crucial role in aging, with overeating increasing the risk of obesity, CVD, and death. Notably, the exceptional longevity of Japanese people living on Okinawa island, a Blue Zone, was partially attributed to mild and constant reduction in calorie intake (64). Smoking, drugs, and alcohol abuse are unequivocally linked to accelerated vascular aging and shorter life expectancy (62,65,66). Nonetheless, moderate alcohol consumption correlates with a lower incidence of CAD and has some cardiometabolic benefits (62). Other factors influencing aging and longevity are air quality, sleep duration and quality, psychological factors, and socioeconomic status (67-71).

DISEASE AND CONDITIONS. Multiple inheritable and acquired conditions and diseases can accelerate vascular aging and lead to an early death. Acquired conditions are frequently caused by an unhealthy lifestyle and are highly interconnected. For instance, obesity is associated with reduced life expectancy, in part due to increased cardiovascular mortality (72). Obesity and fat distribution influence many vascular age measures, including arterial stiffness, carotid IMT, and inflammation markers. Obesity is also a risk factor for type 2 diabetes (T2D), a metabolic disorder featuring hyperglycemia, insulin resistance, and increased CVD-dependent mortality. Moreover, T2D is a common cause of chronic kidney disease, which is characterized by extensive vascular calcification and increased CVD-related mortality (73).

Atherosclerosis is accelerated by autoimmune diseases, such as systemic lupus erythematosus and

rheumatoid arthritis, in accordance with the central role of inflammation in aging and associated vascular disease (74). Furthermore, premature vascular aging is a common presentation in individuals infected with human immunodeficiency virus, in part due to the side effects of antiretroviral drugs (75).

An early onset of vascular aging is a feature of multiple genetic diseases. Familial dyslipidemias, such as familial hypercholesterolemia, are a group of genetic diseases that alter lipid metabolism and require pharmacological treatment to prevent early-onset atherosclerosis (76). Generalized arterial calcification of infancy and pseudoxanthoma elasticum are inherited syndromes associated with defective extracellular pyrophosphate metabolism and arterial calcification (77). Generalized arterial calcification of infancy begins in utero and causes severe calcific stenosis, hypertension, heart failure, and death within the first 6 months of life. Pseudoxanthoma elasticum has a milder course and features CVD. Hutchinson-Gilford progeria syndrome is caused by mutations in the *LMNA* gene and features most vascular alterations found in the elderly, such as atherosclerosis, arterial stiffness, and calcification. These defects lead to death in the second decade of life, typically from myocardial infarction or stroke (1). Patients with Werner syndrome carrying mutations in the *WRN* gene develop age-related diseases, including T2D, atherosclerosis, myocardial infarction, and cancer, and die at an average age of 54 years (78).

STRATEGIES TO DECELERATE VASCULAR AGING

Studies of factors and diseases that accelerate and decelerate biological aging have identified multiple mechanisms that control this process. This section summarizes key behavioral and pharmacological interventions targeting these processes with the potential to promote vascular health and longevity (**Central Illustration, Table 1**).

BEHAVIORAL STRATEGIES. Aside from the obvious measures of avoiding or quitting smoking and avoiding excessive alcohol consumption, several other interventions can reduce both global and vascular biological aging. One of the most prominent measures is physical activity, especially aerobic exercise, which is linked to a reduced incidence of age-related diseases, decreased vascular and molecular measures of biological age and CVD risk factors, and an extended life span in humans (60,61,79–82).

Caloric restriction and fasting are the best documented strategies to prolong the life span across several model species (81). Even without increasing physical activity, reducing energy intake in obese humans improves endothelial function and reduces arterial stiffness and blood pressure, partially due to weight and fat mass loss (82,83). Likewise, 2-year follow-up in the CALERIE (Comprehensive Assessment of Long-term Effects of Reducing Intake of Energy) clinical trial showed reductions in CVD risk (84), inflammation (64), and biological age in nonobese subjects (85). Nevertheless, sustained calorie restriction can lead to the loss of lean muscle mass and bone density (82), and its long-term effects on survival in nonobese humans remain unknown.

Changes in dietary composition can also have a strong modulatory effect on aging and vascular disease. For example, the Mediterranean and Dietary Approaches to Stop Hypertension diets reduce long-term weight gain and are associated with a lower risk of adverse clinical events (62). Likewise, high vegetable and fruit intake is associated with improved endothelial function and decreased arterial stiffness and blood pressure (63,82). Arterial function can be improved and CVD risk reduced by the consumption of cocoa, tea, coffee, fermented dairy products, fish, nuts, seeds, whole grains, legumes, and olive oil (63). Furthermore, dietary sodium restriction lowers blood pressure and reduces arterial stiffness (82).

PHARMACOLOGICAL STRATEGIES. Despite the unquestionable beneficial effects of behavioral strategies on heath span and life span, long-term adherence to these habits is typically low. Thus, pharmacological targeting of pathways that are modified by behavioral approaches presents a more practical alternative. Key signaling cascades regulated by caloric restriction are the energy- and nutrient-sensing pathways, including the mechanistic target of rapamycin (mTOR), adenosine monophosphate–activated protein kinase (AMPK), and sirtuin pathways (5). Aging dysregulates these pathways, and their pharmacological modulation extends life span in various species, including mammals (86,87).

Rapamycin, an inhibitor of mTOR that also activates AMPK, is typically used as an immunosuppressant in organ transplant recipients and as an antiproliferative agent in the treatment of some cancers. Compared with other immunosuppressants, rapamycin diminishes arterial stiffness, blood pressure, and carotid IMT in kidney transplant recipients, suggestive of its vasculoprotective properties (88–90). Furthermore, rapamycin and its analogues (rapalogs) show antiatherosclerotic properties in preclinical models and are used clinically to prevent in-stent restenosis and cardiac allograft vasculopathy (91,92). Nevertheless, the use of rapamycin to

TABLE 1 Behavioral and Pharmacological Strategies to Decelerate Biological Aging of the Vascular System

Strategy	Intervention or Treatment	Beneficial Effects	Side Effects	Ref. #
Behavioral				
	Physical activity	Reduced CVD risk and mortality Loss of abdominal fat and reduction of waist/hip ratio Improved measures of vascular health (endothelial function, arterial stiffness, and blood pressure) Favorable outcomes in the lipid profile Reduced inflammation markers Increased insulin sensitivity	Excessive exercise in the elderly may increase mortality risk Excessive endurance exercise may have adverse effects on cardiac structure and function	(60,81,82,103–105)
	Caloric restriction	Reduced CVD risk Weight and fat mass loss Improved measures of vascular health (endothelial function, arterial stiffness, blood pressure, and carotid intima-media thickness) Reduced cholesterol and TG Reduced inflammation markers Increased insulin sensitivity	May produce loss of bone density and lean muscle mass Risk of malnutrition (in lean humans)	(64,82,83,86)
	Fasting and fasting-mimicking diets	Reduced weight, waist circumference, and abdominal fat Reduced blood pressure Decreased glucose, total cholesterol, LDL, and TG Reduced inflammation markers Increased insulin sensitivity	Fatigue, weakness, and headaches Might be harmful to children, under weight people, the elderly, and type 1 diabetes and extreme hypertension patients	(81,106–109)
	Healthy foods and dietary patterns	Reduced CVD risk and mortality Reduced long-term weight gain Improved measures of vascular health (endothelial function, arterial stiffness, and blood pressure) Favorable outcomes in the lipid profile Reduced inflammation markers Positive effects on gut microbiota composition and diversity		(62,82,110,111)
	Low sodium intake	Improved measures of vascular health (endothelial function, arterial stiffness, blood pressure) Lower risk of cardiovascular events (?)*	Possible negative impact on lipid profile Increased renin, aldosterone, noradrenaline, and adrenaline Orthostatic complaints (in chronic kidney disease patients)	(62,63,82,112,113)

Continued on the next page

promote healthy vascular aging is limited by adverse side effects, including hyperglycemia, hyperlipidemia, and insulin resistance (93). Ongoing research is testing several rapalogs in the search for a safer alternative, and some beneficial effects have been reported, including enhanced immune function and reduced infections in the elderly (94).

Metformin is the most prescribed drug for T2D and shows very mild side effects (87). Apart from increasing insulin sensitivity, metformin targets a series of age-related mechanisms, including AMPK activation and mTOR inhibition. Metformin treatment reduces vascular age measures, including arterial stiffness, endothelial dysfunction, and CACS (95,96) and decreases CVD risk and mortality (87).

Resveratrol is a polyphenol naturally present in red wine, grapes, and other berries. It activates sirtuin-1 and AMPK and inhibits the mTOR pathway (97). Resveratrol has shown vasculoprotective effects in preclinical models, and clinical trials have demonstrated moderately diminished systolic blood pressure in hypertensive patients and glycemia in T2D patients; however, some undesirable side effects have been reported, such as possibly blunting the benefits of exercise in the elderly (98). Nicotinamide adenine dinucleotide precursors, such as nicotinamide riboside and nicotinamide mononucleotide, are another group of sirtuin activators that ameliorate vascular aging in mice (99). Preliminary studies in humans suggested their potential use to reduce arterial stiffness and blood pressure (100), and a clinical trial was launched in 2019 (101).

Strategies targeting inflammation are emerging as potential therapies to counteract vascular aging (82). Blockade of tumor necrosis factor-α reduced arterial stiffness and carotid IMT in rheumatoid arthritis

TABLE 1 Continued

Strategy	Intervention or Treatment	Beneficial Effects	Side Effects	Ref. #
Pharmacological				
	Rapamycin and analogues	Reduced in-stent restenosis and associated risk of major adverse cardiac events	Metabolic defects (hyperglycemia, hyperlipidemia, insulin resistance, and increased incidence of type 2 diabetes)	(81,88-90,92,114-116)
		Delayed progression of cardiac allograft vasculopathy	Mucositis and rash	
		Diminished arterial stiffness, blood pressure, and carotid intima-media thickness (compared with other immunosuppressants in kidney transplant patients)	Severe infections	
			Anemia, thrombocytopenia	
			Proteinuria	
			Impaired wound healing	
	Metformin	Reduced CVD risk and mortality	Minor gastrointestinal side effects	(82,87,115,117,118)
		Weight and fat mass loss	Hypoglycemia (when used with antidiabetes drugs or insulin)	
		Improved measures of vascular health (arterial stiffness, blood pressure, coronary artery calcium score, and carotid intima-media thickness)	Lactic acidosis (when there is renal insufficiency; rare)	
		Reduced total cholesterol, LDL, and TG and increased HDL	Anemia (rare)	
		Reduced inflammation		
		Increased insulin sensitivity		
	Resveratrol	Weight loss and reduced waist circumference and fat mass	Mild to moderate gastrointestinal symptoms (at high doses)	(98,119,120)
		Improved cerebral blood flow	May counteract the cardioprotective effects of exercise	
		Reduced blood pressure (?)		
		Decreased plasma glucose, TG, and total cholesterol (?)		
		Decreased inflammation markers (?)		
	NAD^+ precursors	Decreased blood pressure and arterial stiffness	Painful flushes (at high doses)	(82,121)
		Decreased TG and LDL and increased HDL		
	Antiproinflammatory cytokine therapies	Reduced risk of recurrent CVD events	Increased incidence of fatal infections	(82,102)
		Reduced arterial stiffness and carotid intima-media thickness		

*Dependent on the degree of sodium restriction.
(?) = inconsistent results between studies; CVD = cardiovascular disease; HDL = high-density lipoprotein; LDL = low-density lipoprotein; NAD^+ = nicotinamide adenine dinucleotide; TG = triglyceride.

patients, and inhibition of interleukin-1β diminished the risk of recurrent CVD events in individuals with previous myocardial infarction and elevated C-reactive protein (82,102). However, the use of antiproinflammatory therapies in heathy subjects is limited by the increased risk of fatal infections.

Because gut dysbiosis contributes to inflammaging, the administration of prebiotics and probiotics might provide additional health benefits (15). Furthermore, a series of medications widely prescribed for chronic age-related vascular disease, such as aspirin, statins, and antihypertensive drugs, might also be considered antiaging pharmaceuticals. However, these compounds are not currently prescribed to healthy individuals.

CONCLUSIONS AND PERSPECTIVES

Age-related vascular damage does not only depend on heritable traits but also can be influenced by lifestyle, environment, and accompanying diseases. Therefore, assessments of vascular disease risk to make recommendations for early prevention should be based not on chronological age but rather on biological age. Although multiple biological age biomarkers have been described, the correlation among them is lower than expected (46), suggesting that some estimates of biological age may reflect different aspects of the aging process. Therefore, more research is needed to validate and refine existing aging biomarkers and to identify more accurate and robust aging indicators. Based on the identification of factors and diseases that change the pace of aging, various antiaging strategies have been proposed to promote healthy aging and postpone the appearance of age-related vascular disease. At present, behavioral antiaging approaches are the most promising; however, they present serious challenges due to limited adherence of individuals to regular exercise, long-term dietary recommendations, and other lifelong lifestyle changes. On the other hand, the use of pharmacological approaches to delay aging in healthy

individuals is still controversial due to their potential long-term side effects, which may exceed the benefits. However, what is clear is that antiaging interventions should focus on extending the health span rather than simply prolonging life span. Strategies to promote healthy aging will not only benefit the individual but also reduce the medical, economic, and sociological impact associated with the progressive aging of the world population.

ACKNOWLEDGMENT The authors thank Simon Bartlett for English editing.

ADDRESS FOR CORRESPONDENCE: Dr. Vicente Andrés, Centro Nacional de Investigaciones Cardiovasculares (CNIC), Melchor Fernández Almagro 3, 28029 Madrid, Spain. E-mail: vandres@cnic.es. Twitter: @HamczykMagda, @CNIC_CARDIO.

REFERENCES

1. Hamczyk MR, del Campo L, Andrés V. Aging in the cardiovascular system: lessons from Hutchinson-Gilford progeria syndrome. Annu Rev Physiol 2018;80:27–48.

2. Savji N, Rockman CB, Skolnick AH, et al. Association between advanced age and vascular disease in different arterial territories: a population database of over 3.6 million subjects. J Am Coll Cardiol 2013;61:1736–43.

3. Laurent S, Boutouyrie P, Cunha PG, Lacolley P, Nilsson PM. Concept of extremes in vascular aging. Hypertension 2019;74:218–28.

4. Jylhävä J, Pedersen NL, Hägg S. Biological age predictors. EBioMedicine 2017;21:29–36.

5. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. Cell 2013;153:1194–217.

6. Blackburn EH, Epel ES, Lin J. Human telomere biology: a contributory and interactive factor in aging, disease risks, and protection. Science 2015;350:1193–8.

7. De Meyer T, Nawrot T, Bekaert S, De Buyzere ML, Rietzschel ER, Andrés V. Telomere length as cardiovascular aging biomarker: *JACC* review topic of the week. J Am Coll Cardiol 2018;72:805–13.

8. Bell JT, Tsai PC, Yang TP, et al. Epigenome-wide scans identify differentially methylated regions for age and age-related phenotypes in a healthy ageing population. PLoS Genet 2012;8:e1002629.

9. Heyn H, Li N, Ferreira HJ, et al. Distinct DNA methylomes of newborns and centenarians. Proc Natl Acad Sci U S A 2012;109:10522–7.

10. Hannum G, Guinney J, Zhao L, et al. Genome-wide methylation profiles reveal quantitative views of human aging rates. Mol Cell 2013;49:359–67.

11. Horvath S. DNA methylation age of human tissues and cell types. Genome Biol 2013;14:R115.

12. Marioni RE, Shah S, McRae AF, et al. DNA methylation age of blood predicts all-cause mortality in later life. Genome Biol 2015;16:25.

13. Fransquet PD, Wrigglesworth J, Woods RL, Ernst ME, Ryan J. The epigenetic clock as a predictor of disease and mortality risk: a systematic review and meta-analysis. Clin Epigenetics 2019;11:62.

14. Fuster JJ, Walsh K. Somatic mutations and clonal hematopoiesis: unexpected potential new drivers of age-related cardiovascular disease. Circ Res 2018;122:523–32.

15. Ferrucci L, Fabbri E. Inflammageing: chronic inflammation in ageing, cardiovascular disease, and frailty. Nat Rev Cardiol 2018;15:505–22.

16. Vaiserman AM, Koliada AK, Marotta F. Gut microbiota: a player in aging and a target for anti-aging intervention. Ageing Res Rev 2017;35:36–45.

17. Strazhesko ID, Tkacheva ON, Akasheva DU, et al. Growth hormone, insulin-like growth factor-1, insulin resistance, and leukocyte telomere length as determinants of arterial aging in subjects free of cardiovascular diseases. Front Genet 2017;8:198.

18. Abdullah SM, Defina LF, Leonard D, et al. Long-term association of low-density lipoprotein cholesterol with cardiovascular mortality in individuals at low 10-year risk of atherosclerotic cardiovascular disease. Circulation 2018;138:2315–25.

19. Bulpitt CJ, Rajkumar C, Cameron JD. Vascular compliance as a measure of biological age. J Am Geriatr Soc 1999;47:657–63.

20. Van Bortel LM, Laurent S, Boutouyrie P, et al. Expert consensus document on the measurement of aortic stiffness in daily practice using carotid-femoral pulse wave velocity. J Hypertens 2012;30:445–8.

21. Townsend RR, Wilkinson IB, Schiffrin EL, et al. Recommendations for improving and standardizing vascular research on arterial stiffness: a scientific statement from the American Heart Association. Hypertension 2015;66:698–722.

22. Mitchell GF, Hwang SJ, Vasan RS, et al. Arterial stiffness and cardiovascular events: the Framingham Heart Study. Circulation 2010;121:505–11.

23. Laurent S, Boutouyrie P, Asmar R, et al. Aortic stiffness is an independent predictor of all-cause and cardiovascular mortality in hypertensive patients. Hypertension 2001;37:1236–41.

24. Humphrey JD, Harrison DG, Figueroa CA, Lacolley P, Laurent S. Central artery stiffness in hypertension and aging: a problem with cause and consequence. Circ Res 2016;118:379–81.

25. Poulter NR, Prabhakaran D, Caulfield M. Hypertension. Lancet 2015;386:801–12.

26. Thijssen DHJ, Bruno RM, van Mil ACCM, et al. Expert consensus and evidence-based recommendations for the assessment of flow-mediated dilation in humans. Eur Heart J 2019;40:2534–47.

27. Skaug EA, Aspenes ST, Oldervoll L, et al. Age and gender differences of endothelial function in 4739 healthy adults: the HUNT3 Fitness Study. Eur J Prev Cardiol 2013;20:531–40.

28. de Groot E, Hovingh GK, Wiegman A, et al. Measurement of arterial wall thickness as a surrogate marker for atherosclerosis. Circulation 2004;109 Suppl 1:III33–8.

29. Bauer M, Caviezel S, Teynor A, Erbel R, Mahabadi AA, Schmidt-Trucksäss A. Carotid intima-media thickness as a biomarker of subclinical atherosclerosis. Swiss Med Wkly 2012;142:w13705.

30. Homma S, Hirose N, Ishida H, Ishii T, Araki G. Carotid plaque and intima-media thickness assessed by B-mode ultrasonography in subjects ranging from young adults to centenarians. Stroke 2001;32:830–5.

31. Lorenz MW, Markus HS, Bots ML, Rosvall M, Sitzer M. Prediction of clinical cardiovascular events with carotid intima-media thickness: a systematic review and meta-analysis. Circulation 2007;115:459–67.

32. Inaba Y, Chen JA, Bergmann SR. Carotid plaque, compared with carotid intima-media thickness, more accurately predicts coronary artery disease events: a meta-analysis. Atherosclerosis 2012;220:128–33.

33. Naqvi TZ, Lee MS. Carotid intima-media thickness and plaque in cardiovascular risk assessment. J Am Coll Cardiol Img 2014;7:1025–38.

34. Lanzer P, Boehm M, Sorribas V, et al. Medial vascular calcification revisited: review and perspectives. Eur Heart J 2014;35:1515–25.

35. Kockelkoren R, Vos A, Van Hecke W, et al. Computed tomographic distinction of intimal and medial calcification in the intracranial internal carotid artery. PLoS One 2017;12:e0168360.

36. Wang Y, Osborne MT, Tung B, Li M, Li Y. Imaging cardiovascular calcification. J Am Heart Assoc 2018;7:e008564.

37. Sangiorgi G, Rumberger JA, Severson A, et al. Arterial calcification and not lumen stenosis is highly correlated with atherosclerotic plaque burden in humans: a histologic study of 723 coronary artery segments using

nondecalcifying methodology. J Am Coll Cardiol 1998;31:126–33.

38. McClelland RL, Chung H, Detrano R, Post W, Kronmal RA. Distribution of coronary artery calcium by race, gender, and age: results from the Multi-Ethnic Study of Atherosclerosis (MESA). Circulation 2006;113:30–7.

39. Raggi P, Gongora MC, Gopal A, Callister TQ, Budoff M, Shaw LJ. Coronary artery calcium to predict all-cause mortality in elderly men and women. J Am Coll Cardiol 2008;52:17–23.

40. McClelland RL, Nasir K, Budoff M, Blumenthal RS, Kronmal RA. Arterial age as a function of coronary artery calcium (from the Multi-Ethnic Study of Atherosclerosis [MESA]). Am J Cardiol 2009;103:59–63.

41. Handy CE, Desai CS, Dardari ZA, et al. The association of coronary artery calcium with noncardiovascular disease: the Multi-Ethnic Study of Atherosclerosis. J Am Coll Cardiol Img 2016;9:568–76.

42. The Multi-Ethnic Study of Atherosclerosis-Arterial Age Calculator. Available at: https://www.mesa-nhlbi.org/Calcium/ArterialAge.aspx. Accessed October 28, 2019.

43. Nilsson Wadström B, Fatehali AH, Engström G, Nilsson PM. A Vascular Aging Index as independent predictor of cardiovascular events and total mortality in an elderly urban population. Angiology 2019;70:929–37.

44. Kim S, Myers L, Wyckoff J, Cherry KE, Jazwinski SM. The frailty index outperforms DNA methylation age and its derivatives as an indicator of biological age. GeroScience 2017;39:83–92.

45. Jia L, Zhang W, Chen X. Common methods of biological age estimation. Clin Interv Aging 2017;12:759–72.

46. Belsky DW, Moffitt TE, Cohen AA, et al. Eleven telomere, epigenetic clock, and biomarker-composite quantifications of biological aging: do they measure the same thing? Am J Epidemiol 2018;187:1220–30.

47. Marais GAB, Gaillard JM, Vieira C, et al. Sex gap in aging and longevity: can sex chromosomes play a role? Biol Sex Differ 2018;9:33.

48. Min KJ, Lee CK, Park HN. The lifespan of Korean eunuchs. Curr Biol 2012;22:R792–3.

49. Gagliano-Jucá T, Basaria S. Testosterone replacement therapy and cardiovascular risk. Nat Rev Cardiol 2019;16:555–74.

50. Kolber MR, Scrimshaw C. Family history of cardiovascular disease. Can Fam Physician 2014;60:1016.

51. Sebastiani P, Bae H, Sun FX, et al. Meta-analysis of genetic variants associated with human exceptional longevity. Aging (Albany NY) 2013;5:653–61.

52. Horvath S, Pirazzini C, Bacalini MG, et al. Decreased epigenetic age of PBMCs from Italian semi-supercentenarians and their offspring. Aging (Albany NY) 2015;7:1159–70.

53. Goel A, Maroules CD, Mitchell GF, et al. Ethnic difference in proximal aortic stiffness: an observation from the Dallas Heart Study. J Am Coll Cardiol Img 2017;10:54–61.

54. Hjelmborg JB, Iachine I, Skytthe A, et al. Genetic influence on human lifespan and longevity. Hum Genet 2006;119:312–21.

55. Palinski W. Effect of maternal cardiovascular conditions and risk factors on offspring cardiovascular disease. Circulation 2014;129:2066–77.

56. Fleming TP, Watkins AJ, Velazquez MA, et al. Origins of lifetime health around the time of conception: causes and consequences. Lancet 2018;391:1842–52.

57. Napoli C, D'Armiento FP, Mancini FP, et al. Fatty streak formation occurs in human fetal aortas and is greatly enhanced by maternal hypercholesterolemia. Intimal accumulation of low density lipoprotein and its oxidation precede monocyte recruitment into early atherosclerotic lesions. J Clin Invest 1997;100:2680–90.

58. Milei J, Ottaviani G, Lavezzi AM, Grana DR, Stella I, Matturri L. Perinatal and infant early atherosclerotic coronary lesions. Can J Cardiol 2008;24:137–41.

59. de Nigris F, Cacciatore F, Mancini FP, et al. Epigenetic hallmarks of fetal early atherosclerotic lesions in humans. JAMA Cardiol 2018;3:1184–91.

60. Lee D, Brellenthin AG, Thompson PD, Sui X, Lee I-M, Lavie CJ. Running as a key lifestyle medicine for longevity. Prog Cardiovasc Dis 2017;60:45–55.

61. Lee DC, Pate RR, Lavie CJ, Sui X, Church TS, Blair SN. Leisure-time running reduces all-cause and cardiovascular mortality risk. J Am Coll Cardiol 2014;64:472–81.

62. Mozaffarian D. Dietary and policy priorities for cardiovascular disease, diabetes, and obesity: a comprehensive review. Circulation 2016;133:187–225.

63. LaRocca TJ, Martens CR, Seals DR. Nutrition and other lifestyle influences on arterial aging. Ageing Res Rev 2017;39:106–19.

64. Madeo F, Carmona-Gutierrez D, Hofer SJ, Kroemer G. Caloric restriction mimetics against age-associated disease: targets, mechanisms, and therapeutic potential. Cell Metab 2019;29:592–610.

65. Banks E, Joshy G, Korda RJ, et al. Tobacco smoking and risk of 36 cardiovascular disease subtypes: fatal and non-fatal outcomes in a large prospective Australian study. BMC Med 2019;17:128.

66. Havakuk O, Rezkalla SH, Kloner RA. The cardiovascular effects of cocaine. J Am Coll Cardiol 2017;70:101–13.

67. Provost EB, Madhloum N. Int Panis L, De Boever P, Nawrot TS. Carotid intima-media thickness, a marker of subclinical atherosclerosis, and particulate air pollution exposure: the meta-analytical evidence. PLoS One 2015;10:e0127014.

68. Zhao B, Vo HQ, Johnston FH, Negishi K. Air pollution and telomere length: a systematic review of 12,058 subjects. Cardiovasc Diagn Ther 2018;8:480–92.

69. Domínguez F, Fuster V, Fernández-Alvira JM, et al. Association of sleep duration and quality with subclinical atherosclerosis. J Am Coll Cardiol 2019;73:134–44.

70. Oliveira BS, Zunzunegui MV, Quinlan J, Fahmi H, Tu MT, Guerra RO. Systematic review of the association between chronic social stress and telomere length: a life course perspective. Ageing Res Rev 2016;26:37–52.

71. Karimi M, Castagné R, Delpierre C, et al. Early-life inequalities and biological ageing: a multisystem Biological Health Score approach in Understanding Society. J Epidemiol Community Health 2019;73:693–702.

72. Jordan J, Nilsson PM, Kotsis V, et al. Joint scientific statement of the European Association for the Study of Obesity and the European Society of Hypertension. J Hypertens 2015;33:425–34.

73. Webster AC, Nagler EV, Morton RL, Masson P. Chronic kidney disease. Lancet 2017;389:1238–52.

74. Frostegård J. Atherosclerosis in patients with autoimmune disorders. Arterioscler Thromb Vasc Biol 2005;25:1776–85.

75. Fourie CMT, Schutte AE. Early vascular aging in the HIV infected: is arterial stiffness assessment the ideal tool? Virulence 2017;8:1075–7.

76. Castro Cabezas M, Burggraaf B, Klop B. Dyslipidemias in clinical practice. Clin Chim Acta 2018;487:117–25.

77. Nitschke Y, Rutsch F. Inherited arterial calcification syndromes: etiologies and treatment concepts. Curr Osteoporos Rep 2017;15:255–70.

78. Oshima J, Sidorova JM, Monnat RJ Jr. Werner syndrome: clinical features, pathogenesis and potential therapeutic interventions. Ageing Res Rev 2017;33:105–14.

79. Delaney JA, Jensky NE, Criqui MH, Whitt-Glover MC, Lima JA, Allison MA. The association between physical activity and both incident coronary artery calcification and ankle brachial index progression: the Multi-Ethnic Study of Atherosclerosis. Atherosclerosis 2013;230:278–83.

80. Tucker LA. Physical activity and telomere length in U.S. men and women: an NHANES investigation. Prev Med 2017;100:145–51.

81. de Cabo R, Carmona-Gutierrez D, Bernier M, Hall MN, Madeo F. The search for antiaging interventions: from elixirs to fasting regimens. Cell 2014;157:1515–26.

82. Nowak KL, Rossman MJ, Chonchol M, Seals DR. Strategies for achieving healthy vascular aging. Hypertension 2018;71:389–402.

83. Martens CR, Seals DR. Practical alternatives to chronic caloric restriction for optimizing vascular function with ageing. J Physiol 2016;594:7177–95.

84. Kraus WE, Bhapkar M, Huffman KM, et al. 2 years of calorie restriction and cardiometabolic risk (CALERIE): exploratory outcomes of a multicentre, phase 2, randomised controlled trial. Lancet Diabetes Endocrinol 2019;7:673–83.

85. Belsky DW, Huffman KM, Pieper CF, Shalev I, Kraus WE. Change in the rate of biological aging in response to caloric restriction: CALERIE biobank analysis. J Gerontol A Biol Sci Med Sci 2018;73:4–10.

86. Fontana L, Partridge L, Longo VD. Extending healthy life span—from yeast to humans. Science 2010;328:321–6.

87. Barzilai N, Crandall JP, Kritchevsky SB, Espeland MA. Metformin as a tool to target aging. Cell Metab 2016;23:1060-5.

88. Johnson RW, Kreis H, Oberbauer R, Brattström C, Claesson K, Eris J. Sirolimus allows early cyclosporine withdrawal in renal transplantation resulting in improved renal function and lower blood pressure. Transplantation 2001; 72:777-86.

89. Joannidès R, Monteil C, de Ligny BH, et al. Immunosuppressant regimen based on sirolimus decreases aortic stiffness in renal transplant recipients in comparison to cyclosporine. Am J Transplant 2011;11:2414-22.

90. Silva AL, Fusco DR, Nga HS, et al. Effect of sirolimus on carotid atherosclerosis in kidney transplant recipients: data derived from a prospective randomized controlled trial. Clin Kidney J 2018;11:846-52.

91. Kurdi A, De Meyer GRY, Martinet W. Potential therapeutic effects of mTOR inhibition in atherosclerosis. Br J Clin Pharmacol 2016;82:1267-79.

92. Chih S, Chong AY, Mielniczuk LM, Bhatt DL, Beanlands RSB. Allograft vasculopathy: the Achilles' heel of heart transplantation. J Am Coll Cardiol 2016;68:80-91.

93. Salmon AB. About-face on the metabolic side effects of rapamycin. Oncotarget 2015;6:2585-6.

94. Mannick JB, Morris M, Hockey HP, et al. TORC1 inhibition enhances immune function and reduces infections in the elderly. Sci Transl Med 2018;10:eaaq1564.

95. Agarwal N, Rice SP, Bolusani H, et al. Metformin reduces arterial stiffness and improves endothelial function in young women with polycystic ovary syndrome: a randomized, placebo-controlled, crossover trial. J Clin Endocrinol Metab 2010;95:722-30.

96. Bannister CA, Holden SE, Jenkins-Jones S, et al. Can people with type 2 diabetes live longer than those without? A comparison of mortality in people initiated with metformin or sulphonylurea monotherapy and matched, non-diabetic controls. Diabetes Obes Metab 2014;16:1165-73.

97. Springer M, Moco S. Resveratrol and its human metabolites—effects on metabolic health and obesity. Nutrients 2019;11:143.

98. Breuss JM, Atanasov AG, Uhrin P. Resveratrol and its effects on the vascular system. Int J Mol Sci 2019;20:1523.

99. de Picciotto NE, Gano LB, Johnson LC, et al. Nicotinamide mononucleotide supplementation reverses vascular dysfunction and oxidative stress with aging in mice. Aging Cell 2016;15:522-30.

100. Martens CR, Denman BA, Mazzo MR, et al. Chronic nicotinamide riboside supplementation is well-tolerated and elevates NAD^+ in healthy middle-aged and older adults. Nat Commun 2018; 9:1286.

101. Nicotinamide Riboside for Treating Elevated Systolic Blood Pressure and Arterial Stiffness in Middle-aged and Older Adults. Available at: https://clinicaltrials.gov/ct2/show/NCT03821623. Accessed October 28, 2019.

102. Angel K, Provan SA, Fagerhol MK, Mowinckel P, Kvien TK, Atar D. Effect of 1-year anti-TNF-α therapy on aortic stiffness, carotid atherosclerosis, and calprotectin in inflammatory arthropathies: a controlled study. Am J Hypertens 2012;25:644-50.

103. Shetty AK, Kodali M, Upadhya R, Madhu LN. Emerging anti-aging strategies - scientific basis and efficacy. Aging Dis 2018;9:1165.

104. Rossman MJ, LaRocca TJ, Martens CR, Seals DR. Healthy lifestyle-based approaches for successful vascular aging. J Appl Physiol (1985) 2018;125:1888-900.

105. Zheng G, Qiu P, Xia R, et al. Effect of aerobic exercise on inflammatory markers in healthy middle-aged and older adults: a systematic review and meta-analysis of randomized controlled trials. Front Aging Neurosci 2019;11:98.

106. Brandhorst S, Choi IY, Wei M, et al. A periodic diet that mimics fasting promotes multi-system regeneration, enhanced cognitive performance, and healthspan. Cell Metab 2015; 22:86-99.

107. Wei M, Brandhorst S, Shelehchi M, et al. Fasting-mimicking diet and markers/risk factors for aging, diabetes, cancer, and cardiovascular disease. Sci Transl Med 2017;9: eaai8700.

108. Longo VD, Panda S. Fasting, circadian rhythms, and time-restricted feeding in healthy lifespan. Cell Metab 2016;23:1048-59.

109. Mattson MP, Longo VD, Harvie M. Impact of intermittent fasting on health and disease processes. Ageing Res Rev 2017;39:46-58.

110. Accardi G, Aiello A, Gambino CM, Virruso C, Caruso C, Candore G. Mediterranean nutraceutical foods: strategy to improve vascular ageing. Mech Ageing Dev 2016;159: 63-70.

111. Calder PC, Bosco N, Bourdet-Sicard R, et al. Health relevance of the modification of low grade inflammation in ageing (inflammageing) and the role of nutrition. Ageing Res Rev 2017; 40:95-119.

112. Garofalo C, Borrelli S, Provenzano M, et al. Dietary salt restriction in chronic kidney disease: a meta-analysis of randomized clinical trials. Nutrients 2018;10:732.

113. DiNicolantonio JJ, O'Keefe JH. Salt and hypertension: what do we know? Curr Opin Cardiol 2018;33:377-81.

114. Soefje SA, Karnad A, Brenner AJ. Common toxicities of mammalian target of rapamycin inhibitors. Target Oncol 2011;6:125-9.

115. Piskovatska V, Strilbytska O, Koliada A, Vaiserman A, Lushchak O. Health benefits of anti-aging drugs. Subcell Biochem 2019;91: 339-92.

116. Rodriguez AE, Granada JF, Rodriguez-Alemparte M, et al. Oral rapamycin after coronary bare-metal stent implantation to prevent restenosis: the prospective, randomized Oral Rapamycin in Argentina (ORAR II) study. J Am Coll Cardiol 2006;47:1522-9.

117. Bjornstad P, Schäfer M, Truong U, et al. Metformin improves insulin sensitivity and vascular health in youth with type 1 diabetes mellitus: randomized controlled trial. Circulation 2018;138:2895-907.

118. Adak T, Samadi A, Ünal AZ, Sabuncuoğlu S. A reappraisal on metformin. Regul Toxicol Pharmacol 2018;92:324-32.

119. Gliemann L, Schmidt JF, Olesen J, et al. Resveratrol blunts the positive effects of exercise training on cardiovascular health in aged men. J Physiol 2013;591:5047-59.

120. Sahebkar A, Serban C, Ursoniu S, et al. Lack of efficacy of resveratrol on C-reactive protein and selected cardiovascular risk factors—results from a systematic review and meta-analysis of randomized controlled trials. Int J Cardiol 2015;189: 47-55.

121. Bogan KL, Brenner C. Nicotinic acid, nicotinamide, and nicotinamide riboside: a molecular evaluation of NAD + precursor vitamins in human nutrition. Annu Rev Nutr 2008;28: 115-30.

KEY WORDS antiaging therapy, biological age, biological age biomarkers, chronological age, vascular disease