ESC European Society of Cardiology

Cardiovascular Research (2025) **121**, 1489–1508
https://doi.org/10.1093/cvr/cvaf138

INVITED SPOTLIGHT REVIEW

How to measure and model cardiovascular aging

Luke Spray[1,2], Gavin Richardson[3], Laura K. Booth[3], Judith Haendeler[4,5], Joachim Altschmied[4,5], Daniel I. Bromage [6], Sienna B. Wallis[1], Konstantinos Stellos [3,7,8,9,10], Simon Tual-Chalot [3]*, and Ioakim Spyridopoulos [1,2]*

[1]Translational and Clinical Research Institute, Vascular Biology and Medicine Theme, Faculty of Medical Sciences, Newcastle University, Centre for Life, Newcastle Upon Tyne NE1 3BZ, UK; [2]Cardiology Department, Freeman Hospital, Newcastle upon Tyne NE7 7DN, UK; [3]Biosciences Institute, Vascular Biology and Medicine Theme, Faculty of Medical Sciences, Newcastle University, Centre for Life, Newcastle Upon Tyne NE1 3BZ, UK; [4]Cardiovascular Degeneration, Clinical Chemistry and Laboratory Diagnostics, Medical Faculty, University Hospital and Heinrich-Heine University Duesseldorf, Duesseldorf 40225, Germany; [5]CARID, Cardiovascular Research Institute Düsseldorf, Medical Faculty, University Hospital and Heinrich-Heine University Duesseldorf, Duesseldorf 40225, Germany; [6]King's College London British Heart Foundation Centre of Excellence, School of Cardiovascular Medicine and Sciences, London, UK; [7]Department of Cardiovascular Research, European Center for Angioscience (ECAS), Medical Faculty Mannheim, Heidelberg University, Mannheim, Germany; [8]German Centre for Cardiovascular Research (DZHK), Partner Site Heidelberg/Mannheim, Mannheim, Germany; [9]Helmholtz-Institute for Translational AngioCardioScience (HI-TAC) of the Max Delbrück Center for Molecular Medicine in the Helmholtz Association (MDC) at the Heidelberg University, Heidelberg, Germany; and [10]Department of Medicine, University Medical Centre Mannheim, Heidelberg University, Mannheim, Germany

Received 7 April 2025; revised 30 May 2025; accepted 14 June 2025

Abstract

Most acquired cardiovascular diseases are more common in older people, and the biological mechanisms and manifestations of aging provide insight into cardiovascular pathophysiology. Measuring aging within the cardiovascular system may help to better understand risk profiles for specific individuals and direct targeted preventative therapy. In this review, we explore telomere attrition, cellular senescence, epigenetic modifications, and mitochondrial dysfunction as key molecular mechanisms of aging. These phenomena are associated with cardiovascular disease through endothelial dysfunction and systemic inflammation, which are measurable in clinical practice with a variety of clinical, laboratory, and imaging techniques. Finally, we discuss that the next tools for modelling cardiovascular aging must be capable of incorporating a vast amount of diverse data from a given patient, pointing to recent developments in artificial intelligence and machine learning.

Keywords Aging • Inflammation • Senescence • Cardiovascular disease • Modelling

This article is part of the Spotlight Issue on Ageing.

1. Introduction

Cardiovascular aging is a fundamental process that contributes to frailty and the development of various life-threatening diseases, including hypertension, heart failure (HF), aortic valve disease, and atherosclerosis.[1–3] As life expectancy continues to rise globally, the burden of age-related cardiovascular conditions has become a critical determinant of both healthspan—the period of life spent in good health—and overall lifespan. The increasing prevalence of these conditions highlights the urgent need to understand and mitigate the effects of cardiovascular aging.

The cardiovascular system plays a multifaceted role in maintaining physiological homeostasis. Beyond its primary function of delivering oxygen and essential nutrients to all tissues while removing metabolic waste, it serves as a central regulator of interorgan communication. The intricate network of blood vessels not only supplies vital resources but also facilitates the exchange of biochemical signals between organs, influencing metabolic, immune, and neurological functions. This complex interplay underscores the circulatory system's role as a gatekeeper of healthy aging.

With advancing age, vascular dysfunction, endothelial impairment, and arterial stiffening progressively disrupt cardiovascular homeostasis.[4,5] These changes increase the risk of systemic inflammation, oxidative stress, and impaired tissue perfusion, all of which accelerate biological aging processes. Measuring and modelling cardiovascular aging have therefore become crucial for identifying early markers of decline and developing targeted interventions. In this review, we focus on imaging, biomarker analysis, and computational modelling that could pave the way for personalized strategies to delay or even reverse cardiovascular aging, ultimately promoting longevity and quality of life.

2. Biological basis of cardiovascular aging

2.1 Telomeres and telomerase

Telomeres, located at the ends of chromosomes, consist of tandem repeats of the hexanucleotide sequence TTAGGG and form a higher-order

* Corresponding author. Tel: +44-1912086000, Email: simon.tual-chalot@newcastle.ac.uk (S.T.-C.). Tel: +44-1912418675; fax +44-1912418666, Email: ioakim.spyridopoulos@ncl.ac.uk (I.S.)

structure stabilized by DNA-binding proteins.[6] These structures prevent degradation, recombination, and chromosome fusions as well as the recognition of chromosome ends as DNA double-strand breaks. Telomere attrition, the gradual shortening of telomeres with each cell division, is a fundamental process of aging. It is an inevitable process, which occurs due to the so-called end replication problem, describing the circumstance that the replication machinery cannot fully duplicate the end of linear double-stranded DNA.[7] After a critical limit of cell divisions, this process leads to cellular senescence, a hallmark of aging.[8,9] This telomere erosion is counteracted by the enzyme telomerase, which—as holoenzyme—consists of the catalytic subunit Telomerase Reverse Transcriptase (TERT), the Telomerase RNA Component (TERC), and several accessory proteins.[10]

2.1.1 Telomere length in atherosclerosis and HF

The most easily accessible patient material is blood, which allows the determination of telomere length in circulating cells by various methods.[11] In humans, shortened telomeres in leukocytes have been consistently observed in individuals with coronary artery disease (CAD) and HF.[12–14] Our own studies have shown that telomere shortening occurs uniformly across various haematopoietic compartments, including bone marrow–derived myeloid cells and thymic progenitor cells, suggesting that leukocyte telomere length (LTL) attrition reflects systemic influences such as increased cellular turnover or DNA damage due to inflammation and oxidative stress—common features of aging—rather than being a direct cause of chronic disease .[15,16]

A large-scale meta-analysis involving 14 studies and over 200 000 participants reported a linear inverse association between LTL and CAD risk: each 1 kb increase in telomere length was associated with an approximate 23% reduction in coronary heart disease (CHD) risk.[12] These findings support LTL as a robust biomarker of atherosclerotic cardiovascular risk.

While the association between short LTL and CHD is well established, evidence for a causal relationship remains inconclusive. Mendelian randomization (MR) offers a powerful strategy to address issues of confounding and reverse causation by using genetic variants as proxies for lifelong exposure.[17] A two-sample MR study of over 470 000 individuals demonstrated that genetic determinants of longer telomeres were associated with a modest but statistically significant reduction in CAD risk within European-ancestry populations, though no causal relationship was observed for cerebral or peripheral atherosclerosis.[18]

Scheller Madrid *et al.*[19] further explored this question using MR in a cohort of 290 000 individuals. They examined three single nucleotide polymorphisms (SNPs) associated with reduced telomere length—located in TERT, TERC, and OBFC1. They found modest but statistically significant increases in ischaemic heart disease risk for variants in TERT (RR: 1.04; 95% CI: 1.02–1.06) and OBFC1 (RR: 1.05; 95% CI: 1.03–1.08), whereas the TERC variant, although associated with shorter telomeres, showed no significant association with disease risk (RR: 1.01; 95% CI: 0.99–1.03). Unlike the protein-coding TERT and OBFC1, TERC encodes the RNA template essential for telomerase activity. These findings raise the possibility that telomere length may serve as a downstream marker of diminished telomerase function, rather than being intrinsically pathogenic. Thus, shorter telomeres alone may not be sufficient to drive pro-inflammatory diseases such as atherosclerosis.

In a UK Biobank cohort of 40 459 middle-aged adults, longer LTL was associated with favourable cardiac remodelling parameters on cardiac MRI and a reduced incidence of HF over a median follow-up of 12 years (HR: 0.86 for highest vs. lowest quartile).[13] Similarly, a larger UK Biobank analysis of ~403 000 individuals without pre-existing cardiovascular disease (CVD) found that individuals in the lowest LTL quartile had a significantly higher incidence of sudden cardiac death, coronary events, and HF hospitalizations.[20] Another UK biobank study using MR in over 470 000 participants suggested that shorter telomere length can decrease life span up to 2.5 years.[21] These studies suggest that longer telomeres may confer resilience against structural cardiac decline and support the concept of LTL as a biomarker of biological rather than chronological aging.

At the myocardial level, cardiomyocytes from patients with HF exhibit significantly shorter telomeres than those from healthy controls. Functional studies using patient-derived induced pluripotent stem cell cardiomyocytes revealed that telomere shortening leads to chromatin remodelling and upregulation of the developmental transcription factor Forkhead Box C1. This, in turn, promotes cellular senescence and contractile dysfunction.[22] These findings provide a mechanistic basis for the clinical observation that short telomeres are associated with poor cardiac outcomes, establishing a direct link between telomere attrition and myocardial failure.

2.1.2 Telomerase and mitochondrial function

Telomere maintenance is undoubtedly critical in stem cells, germ cells, and tissues with high proliferative capacity. However, other aging-related mechanisms could be more important in slowly or non-dividing cells, like neurons and the major structural cell types of the cardiovascular system, cardiomyocytes, endothelial cells (ECs), vascular smooth muscle cells, and fibroblasts. Here, mitochondrial dysfunction, another hallmark of aging,[9] might be more relevant. The functions of mitochondria reach far beyond energy provision as they integrate multiple metabolic signals and, thus, serve as a central node in metabolism[23] and signalling organelles.[24,25] Moreover, they are at the center of oxidative metabolism, as they detoxify molecular oxygen in the respiratory chain and themselves are one of the major intracellular producers of reactive oxygen species (ROS).[26] Thus, they play an important role in the cellular redox homeostasis, which becomes disturbed with increasing age resulting in oxidative stress.[27,28] The constant work of cardiomyocytes consumes enormous amounts of adenosine triphosphate (ATP), generated by mitochondria, but ECs, vascular smooth muscle cells, and fibroblasts all also rely on proper mitochondrial function.[29–31] Mitochondrial dysfunction is therefore a typical feature of CVD.[32]

Interestingly, TERT is also critical for normal mitochondrial function. The holoenzyme telomerase was originally identified as a nuclear enzyme responsible for telomere maintenance in the unicellular eukaryote *Tetrahymena*[33] and subsequently also in humans.[34] Nearly two decades later, its catalytic subunit TERT has been detected in mitochondria of human cells by several independent groups.[35–38] Localization of TERT in these organelles can be explained by the presence of a *bona fide* mitochondrial targeting sequence at the N-terminus of the mammalian protein,[35,37] in addition to the nuclear import and export signals,[39,40] allowing transport of TERT into either of the two organelles. However, the mode of action in mitochondria must be different from the nucleus as the circular mitochondrial DNA (mtDNA) does not contain telomeres and because TERC is not imported into mitochondria.[41] A direct link between TERT and mitochondrial functions was originally provided by the observations that TERT binds to mtDNA and protects it against damage,[37,41] reduces mitochondrial superoxide levels[36,37,40,41] and is required for full complex I activity,[37,42] several processes interrelated to each other. Moreover, expression of a TERT mutant defective in mitochondrial import led to ultrastructural changes in mitochondria,[41] while expression of TERT forced into the mitochondria restored nitric oxide (NO)-mediated dilation of microvessels from patients with CAD.[43]

All studies to this point were either performed by expressing TERT forced into the cell nucleus or the mitochondria by disruption or addition of specific targeting signals, on a TERT-proficient cellular background, or in global TERT-knockout animals and organs thereof. This made it difficult to unequivocally assign specific effects to nuclear or mitochondrial TERT, respectively, especially *in vivo*. This dilemma was solved by the generation of two unique mouse models containing TERT exclusively in one of the cellular compartments in all cells of the body.[44] Using these mice, it was shown that mitochondrial, but not nuclear TERT is necessary and sufficient to maintain mitochondrial complex I activity and to ameliorate ischaemia/reperfusion injury of the heart. The latter can be ascribed—at least in part—to protection of cardiomyocytes against apoptosis, improved revascularization, and enhanced myofibroblast differentiation.[44]

Thus, mitochondrial TERT has a protective effect with respect to age-related CVDs, and it may be desirable to increase the levels of TERT

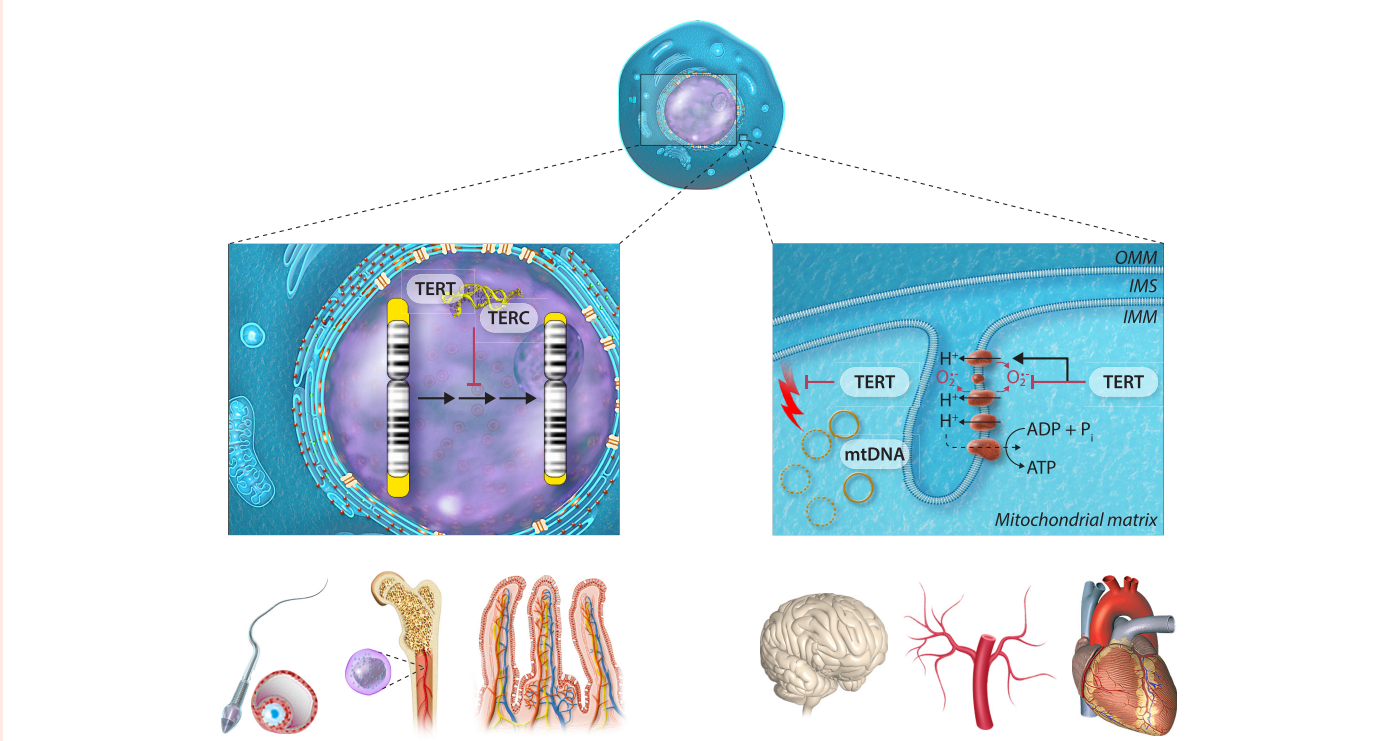

Figure 1 Telomerase reverse transcriptase counteracts different hallmarks of aging through dissimilar, organelle-specific mechanisms. In the nuclei of germline cells, stem cells, e.g. in the bone marrow or in intestinal crypts and of rapidly dividing cells like the intestinal epithelium, the telomerase holoenzyme consisting of TERT, TERC, and accessory proteins prevents telomere erosion and, thus, induction of cellular senescence. Conversely, in slowly or non-dividing cells of e.g. the brain, the vasculature and the heart, mitochondrial TERT protects mitochondrial DNA against damage, maintains activity of respiratory chain complex I and limits superoxide production, thereby maintaining mitochondrial functionality. TERT, telomerase reverse transcriptase; TERC, telomerase RNA component; OMM, outer mitochondrial membrane; IMS, intermembrane space; IMM, inner mitochondrial membrane; mtDNA, mitochondrial DNA.

in mitochondria in humans. Such an effect has been demonstrated with TA-65®, a purified extract from the medicinal plant *Astragalus membranaceous* in primary human ECs, along with improved migratory capacity[44] and reduced ROS release in brain mitochondria.[45] Moreover, dietary restriction increases mitochondrial TERT levels in the brain.[46] In a clinical setting, remote ischaemic preconditioning in patients undergoing coronary artery bypass grafting, which had a cardioprotective effect that was accompanied by improved mitochondrial respiration,[47] also led to an increase in mitochondrial TERT levels in right atrial appendages.[44]

In summary, TERT uses different modalities to counteract at least two hallmarks of aging, namely telomere erosion and mitochondrial dysfunction (*Figure 1*). In rapidly dividing cells, its most important role might be the prevention of telomere erosion. Its predominant effect in cells with low proliferative capacity, such as those in the brain and the cardiovascular system, appears to be the maintenance of mitochondrial function, specifically supporting electron transport chain function and restricting production of ROS.

2.2 Senescence

On a whole-organism level, a recognized feature of aging pathologies is the accumulation of senescent cells: these are defined as cells, which have irreversibly exited the cell cycle and display a particular combination of characteristics including overexpression of pro-survival Bcl-2 family proteins and expression of the pro-inflammatory senescence-associated secretory phenotype (SASP).[48,49] From its initial definition as simply replicative exhaustion,[50] it is now known that cellular senescence is a complex phenotype, which can be acquired in a telomere-independent fashion by post-mitotic cell types, including cardiomyocytes, and can contribute to disease in various organ systems, including atherosclerosis,[51,52] myocardial infarction (MI),[53] and HF.[54,55] In addition to replicative exhaustion, senescence can be induced by a range of stressful stimuli, termed stress-induced premature senescence, with the common features of oxidative stress and mitochondrial dysfunction. Overproduction of ROS, or inadequate antioxidant processes, leads to DNA damage and a DNA damage response (DDR).[56] DDR then leads to cell cycle arrest, through activation of the p53/p21 pathway, and senescence.

Multiple cardiovascular risk factors beyond age are also closely linked with senescence. Hyperglycaemic media shifts cells towards senescence *in vitro*, and senescence of pancreatic islet cells has been implicated in the development of diabetes.[57] High levels of lipids also contribute to senescence as, in the presence of ROS, lipids are oxidized and cause ER-stress.[58] White adipose tissue (WAT) is especially prone to senescence, and this is accelerated in the presence of obesity or diabetes.[59,60] In models of obesity, senescent WAT cells appear very early in the disease process, suggesting senescent cells may contribute to the core pathogenesis of obesity and insulin resistance.[61–63] Cigarette smoking also accelerates senescence,[64] and in mouse models targeting senescence improves smoking-related lung disease.[64] Sarcopenia, the loss of muscle mass which commonly occurs with aging, is also strongly linked to senescence and CVD incidence.[65]

Attenuated macroautophagy and an accumulation of dysfunctional mitochondria via impaired mitophagy are recognized hallmarks of aging,[9] and of cellular senescence.[48,66] Accordingly, short hairpin RNA–based knockdown of key mitophagy components, such as PINK1, Parkin, or p62, is sufficient to induce senescence. Further, several pharmacological interventions identified as 'anti-senescent' have been found to promote

mitophagy, with their anti-senescent effects being dependent on functional expression of the autophagic machinery.[67] In the context of CVD and myocardial aging, mouse models have demonstrated that functional autophagy within cardiomyocytes is critical to maintain myocardial health. Disruption in autophagy via cardiomyocyte-specific deletion of Atg5 results in myocardial dysfunction with characteristics of age-related remodelling.[68] In non-cardiomyocyte cell lineages, including fibroblasts and primary human cardiac microvascular ECs, enhancing autophagy has been shown to have therapeutic potential. Novel small molecules that activate mitophagy machinery, such as p62, have shown promising anti-senescence and anti-aging properties.[67] The action of many longevity-promoting interventions often converges on the activation of AMP kinase (AMPK), activation of the Sirtuin pathway, or dampening of mechanistic target of rapamycin. Some suggest that widespread changes in these complex and far-reaching pathways (such as by administering rapamycin) are regulated in the cardiac microenvironment by post-transcriptional mechanisms such as micro RNA (miRNA) networks.[69,70] Certainly, more precise molecular mechanisms and targeting strategies must be elucidated to maximize the potential of any longevity-promoting intervention. Although tracking changes in mitophagic flux in real time is technically challenging, fluorescent reporters have recently been used both *in vitro* alongside orthogonal studies, and *in vivo*, to great effect.[67,71]

The mechanisms by which enhanced autophagy appears to improve cellular function centre on improved mitochondrial function, as a result of increased mitophapy and mitochondrial turnover. This is perhaps unsurprising, as dysfunctional mitochondria contribute to several pathological processes related to aging. Mitochondria act as a source of oxidants in the cell, and mitochondrial dysfunction (e.g. decreased membrane potential and increased proton leak) accelerates senescence in large part by release of ROS.[72,73] Mitochondria-derived ROS form part of the SASP and studies using mitochondria-depleted senescent cells show that the organelles are in fact required for expression of the SASP; mitochondria-depleted senescent cells remain in cell cycle arrest but fail to express the classical senescence markers p16 and p21, or the typical pro-inflammatory and pro-oxidant phenotype.[74] It has been suggested that increased permeability of the outer mitochondrial membrane in senescent cells allows the release of mtDNA, which can activate the cGAS-STING pathway (normally responsible for recognising exogenous, pathogenic, cytoplasmic DNA).[75,76] Additionally, against the backdrop of increased pro-survival pathway expression, insufficient cytochrome c release and caspase activation in a senescent cell may result in sublethal apoptotic processes, which induce DNA damage and contribute to further genetic instability.

Some argue that nuclear DNA leakage may also be a trigger for cellular DNA sensors and inflammation. Senescent cells have been shown to extrude chromatin fragments from their nuclei to the cytoplasm, thereby triggering the cGAS-STING pathway.[77] Consequently, NF-kB signalling is activated, transcription of pro-inflammatory genes is switched on, and SASP released. Other DNA sensors may also be activated by nuclear or mtDNA in senescent cell cytoplasm, including toll-like receptor 9, and the inflammasome complex. Clinically, there is significant interest in how circulating mtDNA correlates with hypertension,[78] and how cell-free mtDNA is associated with MI.[79] Dampening DNA-leakage sensors using small molecules has been investigated in preclinical studies,[80] and further research is warranted to assess their potential in senescence and CVD.

With aging being the biggest risk factor for CVD, prophylactically or curatively abrogating senescent cell burden in the aging heart is an exciting concept. Landmark studies explored this notion using senolytics: pharmacological agents, such as Navitoclax, which selectively induce senescent cells to apoptosis by inhibiting Bcl-2 family proteins.

Suggesting that senescent cells may be detrimental post-MI, the use of Navitoclax in mice post-MI has been shown to attenuate cardiomyocyte hypertrophy and myocardial profibrotic TGFβ2 expression.[81] Standard treatment of MI involves reperfusion of the ischaemic area of myocardium, but this sudden reperfusion is itself associated with localised oxidative stress and inflammation, termed ischaemia-reperfusion injury (IRI).[82] In an IRI setting, Navitoclax treatment was associated with reduced infarct scar size, increased angiogenesis, and reduced SASP expression.[53,81]

Though the heart is thought to have limited regenerative capacity overall, a small population of cardiac progenitor (or stem) cells (CPCs) are thought to underpin this capacity, which is important for reparative potential post-insult. In patients > 70 years of age, over half of CPCs have been shown to be senescent, and unable to fulfil their regenerative, reparative role in an infarcted heart. Furthermore, the SASP of these CPCs was able to induce senescence in non-senescent CPC populations *in vitro*, but the addition of senolytic combination therapy with dasatinib and quercetin abrogated these effects.[83]

Overall, several aspects of the senescent cell phenotype may lend themselves to promotion of CVD in an aging setting, including the pro-inflammatory SASP and the loss of any limited regenerative potential. The prospect of senolytics is exciting, but the long-term effects of removing cells from a post-mitotic cardiomyocyte population are uncertain. It is heartening, however, that Navitoclax is being employed in Phase II clinical trials in an oncology setting and shows a favourable safety profile. Novel approaches such as senomorphics (drugs which modify or dampen the senescent phenotype, particularly SASP) may hold more promise, but even these show mixed efficacy from preclinical studies.[84] Certainly, a better understanding of the detrimental aspects of the senescent phenotype will allow for more targeted therapeutic approaches.

2.3 Epigenetics

One of the key mechanisms contributing to the chronic low-grade inflammation observed in aging cardiovascular systems is driven by dynamic and flexible age-related epigenetic modifications. These modifications, including DNA methylation, histone modifications, alterations in chromatin structure, and RNA-based mechanisms, play a pivotal role in controlling the gene expression of inflammatory pathways without altering the underlying DNA sequence.[85,86] As aging progresses, epigenetic alterations occur sporadically in response to both exogenous and endogenous factors and are closely associated with healthspan and lifespan.[87–89] Sex-specific differences have also been observed in genome-wide DNA methylation patterns and associations with several cardiometabolic traits and varying risks of CVD, including MI and stroke.[90,91] These findings suggest that the epigenetic landscape might play a critical role in understanding disease phenotypes and tailoring sex-specific treatments. Epigenetic age is therefore emerging as a personalized and accurate predictor of biological age. It has been linked to numerous age-related diseases and mortality,[92] while epigenetic age acceleration is associated with the presence of subclinical atherosclerosis, a process mediated by systemic inflammation.[93] Epigenetic mechanisms can be modified by pharmacological agents, lifestyle interventions, or diet.[94–96] Consequently, epigenetic clocks that track biological age may serve as valuable tools for interventions aimed at mitigating the effects of aging, especially if they can detect biological aging in young individuals who show no signs of disease.

2.3.1 DNA methylation

DNA methylation is a dynamic process involving the addition of a methyl group by DNA methyltransferases (DNMTs) or its removal by ten-eleven translocation methyl-cytosine dioxygenases (TET), primarily targeting cytosine residues in CpG dinucleotide sites, leading generally to transcriptional repression. These enzymes are regulated by genetic and environmental factors and by age.[97,98] DNA methylation influences the inflammatory response of circulating leukocytes, enhances the release of inflammatory cytokines, and promotes the progression of age-related CVDs.[99–102] In the general population, over 10% of people older than 70 years harbor blood cell clones with loss-of-function mutations in epigenetic modifiers such as TET2 and DNMT3A.[103] While 75% of CpG sites are typically methylated in mammalian cells, aging leads to deviations in global DNA methylation patterns. Global hypomethylation occurs alongside localized hypermethylation at specific loci, contributing to genomic instability.[104–107] Atherosclerotic lesions in humans and preclinical models exhibit global DNA hypomethylation, while promoter regions of atheroprotective genes associated with endothelial and smooth muscle cell functions often show hypermethylation.[108–111] Reduced DNA methylation has also been

observed in the promoter region of TNFα,[112] a potent inflammatory cytokine associated with vascular aging.[113,114] Additionally, age-dependent DNA hypomethylation regulates interleukin (IL)-1β and IL-6 expression.[115,116] Since the benefit of IL-1β-targeting therapies like canakinumab depends on the magnitude of the IL-6 response,[117,118] DNA methylation levels may partly explain why patients with somatic variations in TET2 or DNMT3A face an increased risk of major adverse cardiovascular events.[119]

2.3.2 Histone modifications and chromatin remodelling

Aging is associated with specific changes in histone levels and their numerous post-translational modifications, including ubiquitination, which alters chromatin structure and accessibility. This shift from tightly packed heterochromatin to loosely organized euchromatin leads to genomic instability, loss of silencing, and increased transcription of retrotransposons.[120–122] Senescent cells accumulate senescence-associated heterochromatin foci, which silence cell cycle-related genes like E2F target genes.[123,124] Changes in specific histones, such as decreased H3K9me3 levels and increased H4K20m33 and H3S10P levels, contribute to inflammageing, the gradually increasing activation of the immune system through aging.[125,126] Sirtuins, a class of histone deacetylases (HDAC), regulate genes involved in NO signalling, oxidative stress, autophagy, and vascular aging through chromatin remodelling.[127] HDAC inhibitors have been shown to significantly reduce TNF-α-stimulated VCAM-1 expression.[128] Similarly, HDAC9 is linked to increased inflammation in advanced plaques, CAD, and ischaemic stroke.[129–131] HDAC9-deficient mice exhibit reduced atherosclerotic lesions, while macrophage-specific HDAC9 deficiency upregulates histone H3 and H4 acetylation and increases ABCA1 and PPARγ levels, preventing cholesterol efflux.[132]

2.3.3 RNA-based mechanisms

Non-coding RNA profiles, including microRNAs (miRNAs), long non-coding RNAs (lncRNAs), and circular RNAs (circRNAs), are profoundly affected by aging and are associated with all-cause mortality and age-related traits.[133–136] These non-coding RNAs serve as critical regulators of multiple biological processes related to aging.[86,137] Several miRNAs, such as miR-21, miR-146a, miR-155, miR-126, and miR-3a, are implicated in inflammageing.[137] Increased levels of miR-34 and reduced expression of its target gene, SIRT1, have been identified in replicative-senescent human ECs, replicative-senescent human aortic smooth muscle cells, and aged mouse aortas.[138,139] In humans, miR-34 is associated with aortic stiffness, a surrogate marker of arterial aging, and the presence of CAD.[140] Notably, leukocyte-specific deletion of miR-34 mitigates atherosclerotic plaque development and enhances Sirt1 expression in an atherosclerosis mouse model.[140] The lncRNA BACE1-AS has been shown to enhance BACE1 mRNA stability, promoting Aβ formation.[141,142] This lncRNA is associated with accelerated vascular aging and the presence, extent, and incidence of atherosclerosis in humans.[143] Although few studies directly link post-transcriptional regulation to inflammageing, accumulating evidence highlights the critical roles of RNA-binding proteins and RNA modifications in age-related diseases.[144–147] The contribution of these RNA metabolism regulatory processes to inflammageing is poised to become a central focus of scientific research in the years ahead.

2.4 Endothelial dysfunction

Aging significantly impacts the vascular endothelium, the monolayer of ECs lining arteries, veins, and capillaries.[148] ECs serve as gatekeepers, regulating the movement of molecules, nutrients, and immune cells between blood and tissues. In their quiescent state, ECs express factors that prevent leukocyte adhesion, platelet activation, and oxidative stress. However, aging leads to a progressive decline in endothelial function, shifting the endothelium towards a proinflammatory, vasoconstrictive, and prothrombotic state, ultimately increasing the risk of CVDs.[148]

A key factor in endothelial dysfunction is the reduced availability of NO, a vasodilator that regulates vascular tone and inhibits platelet aggregation. The decline in NO bioavailability reduces the ability of blood vessels to expand and contract, contributing to elevated blood pressure.[149,150] The mechanisms underlying age-related endothelial dysfunction also include increased oxidative and nitrosative stress, cellular senescence, mitochondrial dysfunction, and impaired angiogenesis.[148,151–153]

Alongside SIRT1's aforementioned role in RNA-based mechanisms, the sirtuin family members SIRT1 and SIRT3 have been implicated as important players tying together endothelial dysfunction, oxidative stress, mitochondrial dysfunction, and cellular senescence in age-associated CVD. Using novel techniques, it was recently shown that chronic, targeted delivery of the phenolic antioxidant compound esculetin to the mitochondria of human aortic ECs resulted in improved mitochondrial respiration and delayed senescence-like features through SIRT1 activation.[154] Furthermore, chronic treatment with this targeted therapy alleviated age-associated atherosclerosis in *Apoe*$^{-/-}$ mice. In aortic ECs, a reported contributing mechanism of esculetin's beneficial effects is enhanced mitochondrial biogenesis via the AMPKα-SIRT3 axis but crucially, targeted delivery of esculetin to the mitochondria is required for its *in vivo* beneficial effect.[155] Altogether, these studies highlight SIRT1 and SIRT3 as common players within interconnecting processes of CVD and aging, whilst also emphasizing the power of targeted, mechanistically-informed interventions like antioxidants.

With aging, ECs show increased expression of adhesion molecules promoting leukocyte recruitment to the arterial wall, a critical step in atherosclerosis development.[156,157] Age-related alterations in cytokine levels, such as IL-6 and TNF-α, further drive inflammation and endothelial activation.[158] Endothelial dysfunction is also linked to metabolic disorders such as diabetes and obesity,[159,160] which worsen insulin resistance and increase the risk of vascular complications.[161] The chronic inflammation associated with impaired endothelial function contributes to metabolic imbalances, providing a fertile ground for the development of age-related diseases.[162,163]

Structural changes in the arterial wall are also observed with aging, including luminal enlargement, intima and media thickening, and medial calcification. Smooth muscle cells undergo phenotypic switching, transforming into a synthetic, osteogenic, and pro-inflammatory phenotype.[149,164] This shift alters the extracellular matrix (ECM) composition, increasing collagen and reducing elastin.[165] Additionally, aged endothelium shows increased permeability,[166,167] allowing infiltration of immune cells that produce ECM-degrading enzymes, such as matrix metalloproteinases.[168] Medial arterial calcification leads to the precipitation of hydroxyapatite crystals in the arterial wall,[169] further contributing to arterial stiffness, an independent predictor of incident CVD and all-cause mortality.[170]

In large arteries, endothelial dysfunction, combined with vascular wall remodelling and calcification, promotes atherosclerosis, a chronic inflammatory disease.[4,171,172] Atherosclerotic lesions typically develop in areas of disturbed blood flow, damaging vascular ECs and triggering inflammation. Compromised endothelial integrity facilitates the accumulation of oxidized low-density lipoprotein particles, leading to monocyte differentiation into macrophages and foam cell formation, contributing to the plaque formation. This may lead to increased vascular resistance and platelet aggregation, raising the risk for hypertension, thrombosis, and acute cardiovascular events, such as MI and stroke.[4,171,172] The relationship between endothelial dysfunction and atherosclerosis appears bidirectional, indicating that these pathological mechanisms exacerbate each other.[173,174] Although aging-associated processes, such as endothelial dysfunction and media remodelling, equally affect veins and arteries, their effects in the two are dramatically different because of the haemodynamic differences. Indeed, human veins do not develop atherosclerosis, but the slower blood flow predisposes to thrombosis, especially in the lower limbs.[175,176] In microvascular beds, aging impairs endothelial vasodilation, endothelial permeability, and reduces capillary density, to compromise the arterial myogenic tone, a mechanism of autoregulation, to maintain a relatively constant blood flow in the capillary bed.[177] Aging-related changes in this process explain why older adults are more prone to hypertension-related complications, such as chronic kidney disease.

Understanding the complex interplay of mechanisms underlying endothelial dysfunction in aging offers promising insights into interventions

that may slow or even reverse these processes. Regular physical exercise has been shown to improve NO production and maintain vascular elasticity.[178] Dietary interventions, like the Mediterranean diet, can mitigate oxidative stress and inflammation.[179] Pharmacological strategies, including cholesterol-lowering and blood pressure-lowering drugs, can further improve endothelial function.[180,181] Similarly, KCa channel activator improved endothelium-dependent vasodilation, and prevented the aging-associated declines in cardiac ejection fraction.[182] Senolytics, which remove senescent ECs *in vitro*, improve cardiac and endothelial function in aged mice.[183,184] By targeting the cellular and molecular mechanisms driving endothelial aging, these interventions may delay or even reverse vascular dysfunction, ultimately reducing the burden of age-related CVD.

2.5 Oxidative stress

Oxidative stress, characterized by an imbalance between the production of ROS and the antioxidant systems that detoxify them, plays a central role in the accelerated progression of cardiovascular aging.[185] This imbalance is particularly detrimental to the vascular endothelium, where excessive ROS generation impairs NO bioavailability, promotes inflammation, and accelerates endothelial dysfunction. Mitochondria, the primary source of ROS in cardiovascular cells, become progressively dysfunctional with age, contributing to a vicious cycle of oxidative damage and cellular senescence.[186,187] Mitochondrial ROS not only damage DNA, proteins, and lipids but also activate redox-sensitive signalling pathways, further exacerbating vascular inflammation and apoptosis.[188] In ECs, this oxidative burden reduces angiogenic capacity, impairs vasodilation, and increases vascular stiffness, which are precursors to age-related CVDs.[189]

The oxidative stress theory of aging, first proposed by Denham Harman in the 1950s, posits that accumulated ROS generated during normal aerobic metabolism cause damage to proteins, lipids, and DNA, thereby accelerating the aging process.[190] Experimental evidence from genetically modified mouse models with altered expression of superoxide dismutase (Sod1 or Sod2), two key antioxidant enzymes, has provided compelling support for this theory. Sod1 knockout (KO) mice, lacking cytosolic superoxide dismutase, display elevated oxidative stress and an accelerated aging phenotype, characterized by muscle atrophy, weakness, and a 30% reduction in lifespan.[191–193] Knockout of mitochondrial superoxide dismutase (Sod2) results in neonatal lethality due to dilated cardiomyopathy, supporting the essential role of mitochondrial ROS detoxification in cardiac development and survival.[194] Heterozygous Sod2+/− mice, which survive into adulthood, exhibit age-dependent endothelial dysfunction, increased mitochondrial oxidative stress, and DNA strand breaks, further linking oxidative stress to vascular aging.[195] Similarly, deficiency of the antioxidant enzyme glutathione peroxidase-1 in aged mice exacerbates vascular inflammation, characterized by monocyte and macrophage infiltration, oxidative DNA damage, and impaired endothelial function.[196] In humans, patients with CAD and low red-cell GPx-1 activity are independently at higher risk of cardiovascular events,[197,198] further highlighting the importance of endogenous antioxidant systems in maintaining vascular integrity during aging.

Importantly, oxidative damage extends beyond local vascular effects, contributing to systemic aging through genomic instability. Levels of oxidized DNA bases such as 8-oxo-2'-deoxyguanosine (8-oxo-dG) have been shown to inversely correlate with lifespan across species,[199] suggesting that the efficiency of DNA repair and antioxidant defences may critically determine species-specific aging rates. The relevance of oxidative stress in premature vascular aging is further supported by studies of Hutchinson–Gilford progeria syndrome (HGPS), a genetic disorder characterized by accelerated aging. ECs derived from HGPS patients exhibit premature senescence, including telomere shortening, increased ROS production, and impaired angiogenic function.[200–202] In hypertensive heart disease, oxidative stress accelerates telomere shortening, and elevated markers of telomeric damage serve as strong predictors of HF progression,[203] confirming that ROS-induced genomic instability may mechanistically link molecular aging to clinical cardiovascular outcomes.

Oxidative stress also interacts with lifestyle and environmental factors that modulate cardiovascular aging.[204] Interventions that reduce oxidative load, such as antioxidant-rich diets, regular physical activity, and pharmacological agents, have shown beneficial effects in both preclinical models and clinical studies. Empagliflozin, a sodium-glucose cotransporter 2 (SGLT2) inhibitor, has been shown to improve endothelial function and reduce mitochondrial oxidative stress in frail hypertensive and diabetic patients, highlighting a potential therapeutic strategy for mitigating vascular aging.[205] Angiotensin II–induced upregulation of SGLT1 and SGLT2 promotes endothelial senescence and dysfunction via oxidative stress pathways, an effects that can be reversed by gliflozins treatment.[206] Metformin, a widely used antidiabetic drug, has also been shown to extend healthspan by reducing oxidative stress, improving endothelial function, and modulating metabolic pathways.[207] Taurine, a sulfur-containing amino acid with antioxidant properties, has been associated with improved endothelial function and reduced oxidative stress in aging models, suggesting its potential as a dietary supplement to counteract cardiovascular aging.[208] Although the direct link to human aging remains to be fully clarified, these findings suggest that targeting oxidative stress pathways may offer benefits against age-related vascular decline.

While the role of oxidative stress in aging is multifaceted, mounting evidence indicates that it acts not only as a marker of biological aging but also as a driver of pathological cardiovascular changes. Thus, targeting oxidative mechanisms presents a promising strategy to delay the onset of CVDs and extend healthspan. However, it is essential to distinguish between physiological ROS signalling, which is crucial for normal vascular tone, immune defence, and cellular adaptation, and pathological oxidative stress, which overwhelms compensatory mechanisms. Future research must therefore focus on refining therapeutic approaches that restore redox balance without impairing vital cellular signalling pathways.

2.6 Inflammation and cardiovascular aging

Inflammageing refers to the increasing activation of the immune system through repeated antigenic stimulation through life.[209] The aged immune system is simultaneously less effective at preventing and clearing infections[210] and overactivated with higher circulating levels of cytokines and higher incidences of some autoimmune diseases, such as giant cell arteritis.[211–213] By helping to unravel this seeming paradox, inflammageing provides insights into many CVDs of aging.

With aging, the composition of the immune cell repertoire changes. Lymphoid-biased haematopoietic stem cells (HSCs) persist in smaller numbers than myeloid-biased HSCs, leading to a myeloid shift in circulating immune cells.[214] This phenomenon can be easily observed in the blood count of older individuals as a higher neutrophil-to-lymphocyte ratio (NLR), which correlates with both chronological age and markers of biological age, such as reduced grip strength.[215] NLR also predicts cardiovascular outcomes, including cardiovascular mortality, in a range of clinical situations, suggesting this myeloid shift may be actively involved in CVD.[216,217] MR analysis of a UK biobank cohort, however, has not found evidence for a causal link between NLR and CAD or MI.[218]

The causative association between systemic inflammation and CVD is well established, through both observational studies and clinical trials. C-reactive protein (CRP) levels predict long-term adverse cardiovascular outcomes similarly well, or better, than cholesterol levels,[219] and inflammatory diseases such as rheumatoid arthritis and systemic lupus erythematosus are robustly associated with increased CVD risk.[220] Systemic inflammation is also common in patients with atherosclerotic CVD (ASCVD), with a large study in Sweden finding 60% of patients with ASCVD have a CRP of 2 mg/L or higher. The inflammatory hypothesis of atherothrombosis was proven with the Canakinumab Anti-Inflammatory Thrombosis Outcome Study, in which the IL-1β inhibitor canakinumab was superior to placebo with regards to cardiovascular events in the secondary prevention setting—the first time an anti-inflammatory medication has been proven effective in improving cardiovascular outcomes. Later, trials of colchicine, another anti-inflammatory treatment, have further cemented the finding that changes in the immune system have a causative relationship with cardiovascular outcomes.[221–223]

Mitochondrial function, as an upstream regulator of inflammation, also represents a promising target for immunomodulatory therapies in CVD. In a randomized clinical trial of 90 post-MI patients aged over 65, twice-daily treatment with the mitochondrial telomerase activator TA-65 for 12–15 months resulted in reduced circulating inflammatory markers and increased numbers of adaptive immune cells compared with placebo.[224] Patients receiving TA-65 also experienced 30% fewer adverse events, suggesting a therapeutic advantage over conventional anti-inflammatory agents, which are often limited by systemic side effects.

In summary, the aging immune system changes substantially, both through a myeloid shift of immune cells and cellular senescence, with SASP-induced inflammation. Inflammation is concretely associated with CV disease, and this inflammation of aging is a key mechanism for adverse CV outcomes in the elderly.

3. Complications associated with cardiovascular ageing

Cardiovascular aging has a wide range of clinical consequences, and most acquired CVDs are linked to aging processes (summarized in *Figure 2*). Aging of large arteries is linked to increased arterial stiffness, hypertension, atherosclerosis, and aneurysm formation—this may result in myocardial ischaemia, thromboembolic events, and spontaneous dissection or rupture of the aorta, all of which carry a risk of fatal consequences.[4] At the core of most aging-related CVDs lies the progressive dysfunction of the endothelium, which impacts arterioles and the microcirculation by impairing EC-dependent vasodilation[225] and promoting microvascular rarefaction.[226] The resulting myocardial hypoperfusion contributes to cardiomyocyte apoptosis and necrosis, which in turn accelerates hypertrophy of surviving cardiomyocytes and stimulates fibroblast proliferation, leading to further left ventricular (LV) hypertrophy.[227]

The loss of arterial myogenic tone[228] in response to increased intraluminal pressure in older individuals heightens the risk of hypertension-related complications, including chronic kidney disease and stroke.[229] Additionally, age-related capillary depletion is linked to vascular cognitive impairment,[228] peripheral artery disease,[230] and macular degeneration.[231,232] In the myocardium, the relatively stable capillary network becomes dysfunctional, increasing the risk of MI with nonobstructive coronary arteries.[233]

Among structural heart diseases, calcific aortic valve disease (CAVD) with haemodynamically significant aortic stenosis is particularly prevalent in older individuals.[234] Although its pathophysiology remains largely unclear, CAVD shares features with arterial stiffness and atherosclerosis,[235] including genetic predisposition, immune cell infiltration, unresolved inflammation, vascular smooth muscle cell phenotypic shifts, and phosphate-calcium metabolism dysregulation.[236]

HF is a leading cause of morbidity in aging populations,[237] characterized by myocardial stiffening, LV thickening, and reduced β-adrenergic receptor

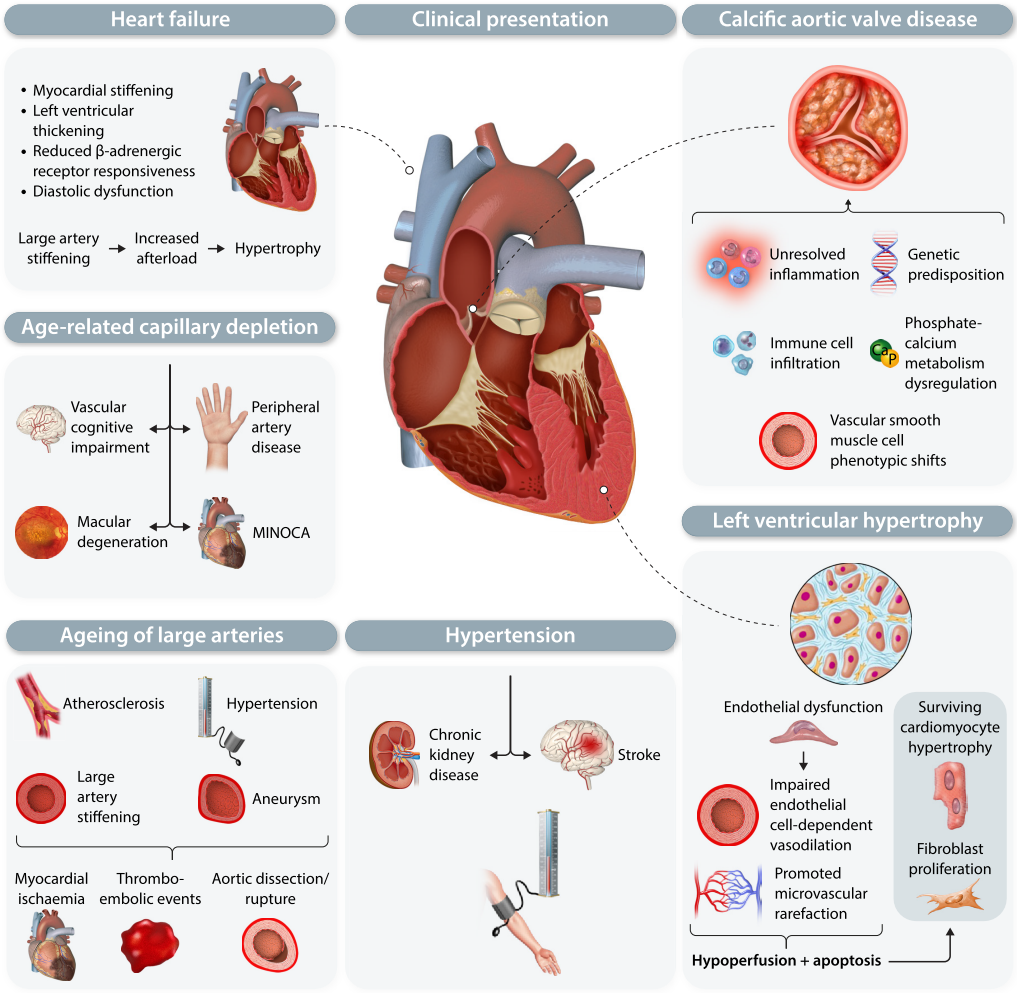

Figure 2 A summary of age-related complications in the cardiovascular system. MINOCA, myocardial infarction with non-obstructive coronary arteries.

responsiveness.[238–241] Large-artery stiffening increases LV afterload, leading to compensatory LV hypertrophy, which in turn raises myocardial oxygen demand.[235,242] Although some studies suggest that aging reduces systolic contractility,[243] diastolic dysfunction is more prevalent, as evidenced by the predominance of HF with preserved ejection fraction (HFpEF) over HF with reduced ejection fraction (HFrEF) in older adults.[244] HFpEF, often secondary to ischaemic insults, is a key contributor to cardiovascular mortality, with diastolic dysfunction emerging as a hallmark of myocardial aging.[245]

The brain vasculature plays a critical role in maintaining cerebral homeostasis, by delivering oxygen and nutrients and removing waste products.[246] Aging of the cerebrovascular system is associated with reduced elasticity of blood vessels, endothelial dysfunction, and decreased perfusion capacity.[247] These age-associated vascular alterations not only impair nutrient and oxygen delivery but also compromise the blood–brain barrier, increasing susceptibility to neuroinflammation and neurodegeneration.[247] Indeed, microvascular impairment and inflammation promote vascular cognitive impairment.[228,248,249] Consequently, compromised cerebral blood flow, microvascular integrity, and vascular remodelling processes have been increasingly recognised as central contributors to age-related neurodegenerative conditions, particularly vascular dementia and Alzheimer's disease.[250,251] These conditions are particularly prevalent in the aging population and are frequently associated with cardiovascular risk factors such as hypertension, atherosclerosis, diabetes mellitus, obesity, and hyperlipidemia.[252–256] Reciprocally, the presence of established cardiovascular risk factors predicts faster cognitive decline.[257] As such, cognitive impairment is tightly connected to cardiovascular aging in a likely bidirectional manner.[258] Importantly, these conditions do not act in isolation but interact synergistically with genetic predispositions and lifestyle factors to influence cognitive trajectories in aging populations.[259] Given these multifaceted interactions, monitoring and managing vascular health, through lifestyle intervention, pharmacological treatment of cardiovascular risks, and early imaging biomarkers, may offer promising avenues for preventing or delaying the onset of vascular cognitive impairment.

4. Measuring cardiovascular ageing

4.1 Clinical biomarkers

Hypertension is among the strongest predictors of incident CVD, and is closely tied to aging.[260] Broadly, hypertension can be classified as either isolated diastolic (IDH), isolated-systolic (ISH), or systolic-diastolic (SDH), depending which of the systolic and diastolic blood pressure (BP) are above the reference limit. Data from the National Health and Nutrition Examination Survey (NHANES), a large population study in the United States, demonstrate that ISH and SDH predict an increased risk of cardiovascular events, while IDH does not.[261,262] Older patients in this study had a remarkably different profile of hypertension. The prevalence of IDH decreased steadily from 39.2% in patients under 40 years old to 0.2% in those over 80, while the prevalence of ISH peaks in the 7th and 8th decades, largely due to arterial stiffness.[263] It is well established that BP then starts to fall in the very old, and that lower BP in the elderly predicts all-cause mortality and even cardiovascular events.[264–266] This makes BP a complex marker of aging in the cardiovascular system, with the trend of a person's BP over many years giving important prognostic information.

Multiple genome-wide association (GWA) studies have sought a heritable basis for extreme longevity, and the SNP rs429358 [apolipoprotein E (ApoE) ϵ4] is consistently associated with lower odds of longevity, while other ApoE variants (ϵ2 and ϵ3) are associated with greater odds.[267,268] ApoE is the major carrying molecule for lipids, and its consistent association with longevity highlights the major impact of atherosclerotic disease on lifespan. ApoE ϵ4 may also hold promise as a novel biomarker, particularly for Alzheimer's dementia.[269,270]

Markers of systemic inflammation, including C-reactive protein (CRP), IL-6, and IL-1β increase steadily with age. These markers are also robustly associated with CVD. CRP is associated with arterial stiffness[271,272] and carotid calcification,[273] although interestingly not with coronary artery calcium (CAC) score.[274] IL-6 and IL-1β, which are upstream of CRP in the same axis, also associate with subclinical and clinical atherosclerosis.[275] Interestingly, MR studies show that genetically lower CRP does not reduce CVD outcomes, while lower IL-6 does.[276] Conversely, polymorphisms which increase the level of the IL-1 receptor antagonist, IL-1R, and so decrease the activity of IL-1, are associated with higher incidence of coronary heart disease.[277] These data suggest that IL-6 may be the key signalling molecule in this pathway, and uniquely amenable to treatment. Clinically, CRP is currently the only inflammatory biomarker for CVD recommended by international guidelines, with the American Heart Association (AHA) recommending its measurement for more detailed assessment of cardiovascular risk.[278]

4.2 The blood-based peptide amyloid-beta 1–40

A new marker of biological age holding great potential is the amyloid-beta 1-40 (Aβ40) blood-based peptide, which is linked to several CVDs (*Figure 3*). Aβ is a proteolytic fragment of the amyloid precursor protein (APP), known for its involvement in Alzheimer's disease.[142] APP is produced in neurons, platelets, cardiomyocytes, and all vascular cells.[141,142] β-secretase (BACE1) is involved in APP cleavage, and further cleavage by γ-secretases generates peptides of length 40 (Aβ40), which is found in vascular lesions, and 42 (Aβ42), which is associated with brain lesions in Alzheimer's disease. The BACE1 antisense transcript (BACE1-AS), a conserved long noncoding RNA, has been found to enhance BACE1 mRNA stability and thus promote Aβ formation.[279,280] Several factors, including inflammation, renal dysfunction, or ischaemia, increase circulating levels and subsequent tissue deposition of Aβ by augmenting its production and processing or by decreasing Aβ clearance.[141,142] Under normal conditions, equilibrium exists between Aβ production and removal. Deregulation of this balance may lead to CVD-associated accumulation of Aβ in the blood, vessels, and heart.[141,142] Increased APP processing and Aβ production may be directly linked to endothelial dysfunction in cerebral and peripheral blood vessels. Aβ peptides at high concentrations are toxic to brain and peripheral ECs, causing cellular damage, enhanced vasoconstriction, and impairment of endothelium-dependent relaxation, thereby promoting atherosclerosis, an age-related disease.[281] Additionally, APP and Aβ have been detected in human carotid plaques and atherosclerotic aortas.[282] Overexpression of APP accelerates atherosclerosis,[283] while APP deletion partially protects against the development of aortic atherosclerosis in ApoE$^{-/-}$ mice.[284] Clinical and experimental evidence indicates that Aβ may play a crucial role not only in the brain but also in the general vasculature. Amyloid deposits are found in the aortic walls of almost 100% of individuals over 50 years of age.[285] Interestingly, the aortas of elderly individuals with either mild fatty streaks or advanced atherosclerotic lesions predominantly contain Aβ40 peptides.[286,287] Furthermore, elevated circulating amyloid concentrations in obesity and diabetes promote vascular dysfunction.[288] Although the source of elevated plasma Aβ40 levels in aged humans remains unknown, endothelial APP significantly contributes to blood Aβ levels, as shown in mice.[289] Unexpectedly, it has been recently proposed that upregulation of APP in the vascular endothelium of aging mice may be an adaptive response designed to protect endothelial function.[290] Altogether, these data suggest that while endothelial Aβ production is necessary for normal function, an excess could contribute to age-related arterial stiffening.

Increased plasma Aβ40 levels have been associated with subclinical cardiac disease, as indicated by elevated high-sensitivity cardiac troponin T, N-terminal pro-B-type natriuretic peptide, and lower LV ejection fraction.[291,292] Additionally, plasma levels of Aβ40 are associated with declining cardiorespiratory fitness in patients without clinically overt CVD.[291] Elevated circulating Aβ40 levels have also been observed in patients with hypertension, diabetes mellitus, and dyslipidemia.[293] In a prospective study of healthy young to middle-aged adults, changes in plasma Aβ40 levels were found to independently predict changes in aortic stiffness, a surrogate marker of vascular aging.[291] In patient at risk for ASCVD, Aβ40 is associated with all-cause mortality partly mediated through renal

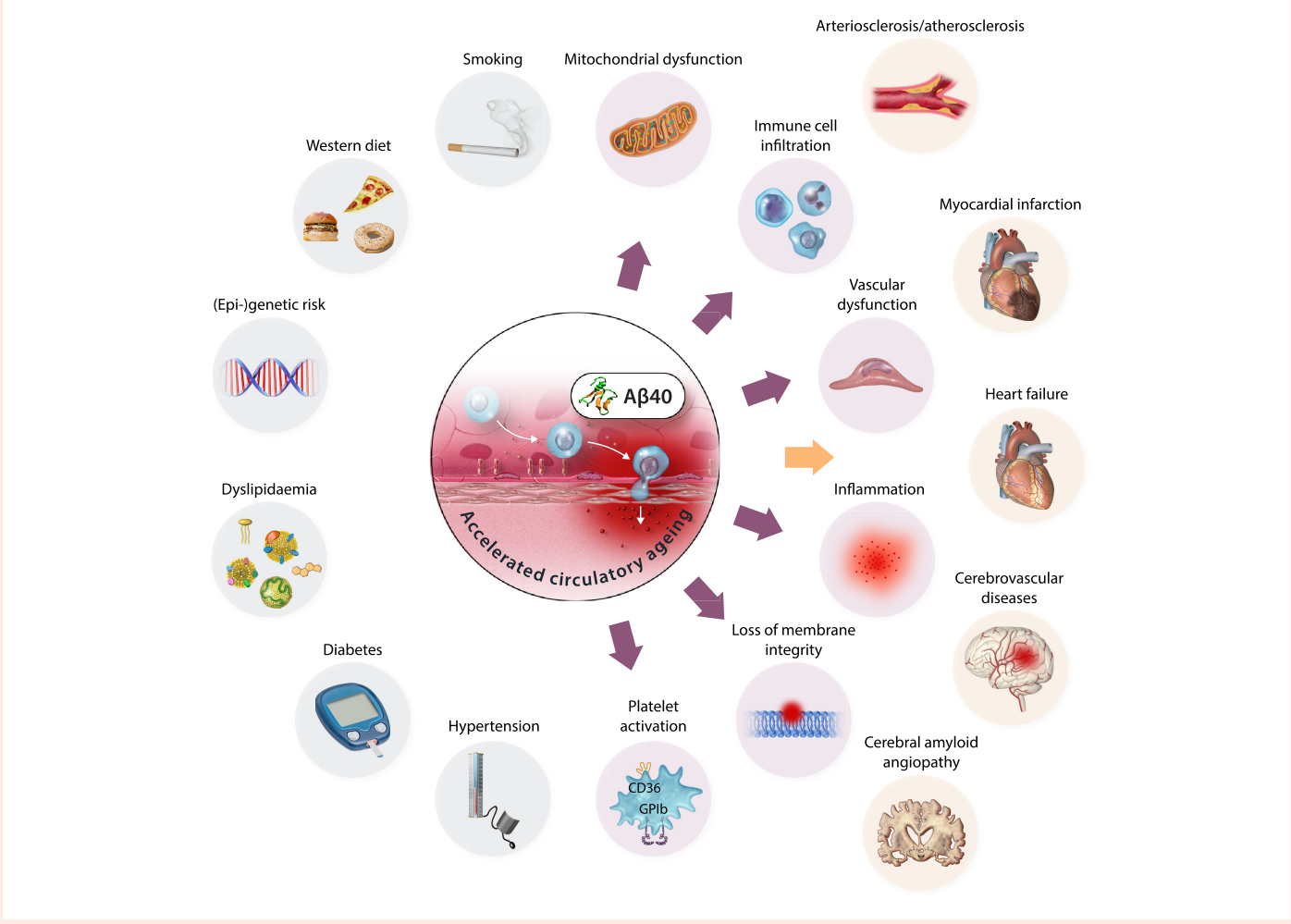

Figure 3 Central role of amyloid-β 1-40 in vascular aging and its contribution to both cardiovascular and neurovascular diseases. Lifestyle and genetics factors, such as a Western diet, smoking, genetic predisposition, dyslipidemia, diabetes mellitus, and hypertension contribute to accelerated cardiovascular aging. The subsequent elevated circulating levels of Amyloid-β 1-40 leads mitochondrial dysfunction, immune cell infiltration, vascular dysfunction, inflammation, loss of membrane integrity, and platelet activation, ultimately promoting arteriosclerosis, myocardial infarction, heart failure, cerebrovascular diseases, and cerebral amyloid angiopathy. Amyloid-β 1-40 as a central driver of the interplay between metabolic, inflammatory, and vascular pathways emerges as a valuable biomarker of cardiovascular aging. Figure created with Biorender: https://BioRender.com/yogrvis.

dysfunction.[294] Aβ40 levels are associated with the presence, extent, and progression of carotid atherosclerosis in postmenopausal women[295] and are linked to plaque composition and burden in patients without clinically overt atherosclerotic CVD.[296] Circulating Aβ40 provides incremental prognostic value and enhances risk stratification in patients with stable CAD and non-ST elevation acute coronary syndrome for predicting adverse cardiovascular events.[297,298] It is also independently associated with mortality in HF patients, possibly due to worsening cardiac function.[292,297,298] Its prognostic significance has largely been demonstrated through retrospectively analyzed prospective studies. Implementing bedside Aβ40 measurement tests in clinical trials could facilitate the establishment of baseline levels and thresholds for predicting adverse outcomes across various age groups. Notably, numerous effective anti-aging strategies have been found to enhance Aβ40 metabolism, highlighting its pivotal role in the aging process.[141]

4.3 CHIP

Clonal haematopoiesis (CH) describes the process when haematopoietic stem and progenitor cells (HSPCs) acquire mutations in genes known to be associated with haematological malignancy but in the absence of an overt blood disorder. These mutations are passed on to progeny cells, resulting in clones of mutant cells that are detectable in the peripheral circulation. CH is defined when the variant allele frequency (VAF) is ≥1% and has been associated with several age-related CVDs. These include atherosclerosis, ischaemic heart disease, MI, HF, as well as outcomes from CVD, including death.[299–303]

CH is an age-related phenomenon as mutations are acquired over time, affecting ~10% of those over the age of 65 years in population studies.[304] Age is also strongly associated with the presence of CH in most reported disease cohorts, as well as being a predictor of accelerated clonal growth.[305] The most frequently identified mutations are in *DNMT3A* and *TET2* genes, genes which encode enzymes responsible for methylating and demethylating DNA CpG sites, respectively. *DNMT3A*- and *TET2*-CH confers abnormal function to mature blood cells, leading to predominantly pro-inflammatory effects.[300,301]

Other than age, clonal growth is also promoted by several established, cardio-metabolic risk factors, including atherosclerosis,[306] obesity,[307,308] and smoking.[309] It is also associated with metabolic dysfunction, specifically low HDL cholesterol levels, which was not observed in patients who

underwent bariatric surgery.[308] CH progression has also been potentiated in a mouse model of obesity, which was partially recovered by anti-inflammatory treatment.[307] Similarly, atherosclerosis has been shown to exacerbate stem cell division and clonal expansion,[306] while smoking was shown to be a causal risk factor for CH in MR studies performed in the UK Biobank.[309] Interestingly, this study also identified longer LTL as a causal risk factor for CH. Given the association between telomere shortening and cellular senescence,[310] this suggests a complex relationship between cellular aging and clonal expansion.

It is therefore not known to what extent CH contributes to or is a consequence of aging. The first human evidence of causality has emerged from the Progression of Early Subclinical Atherosclerosis (PESA) study.[311] Based on longitudinal CH assessment and serial vascular imaging, this study suggested that CH has a unidirectional, causative association with the development of atherosclerosis. Specifically, having a mutation related to CH, especially at higher VAF, was associated with an increased risk of developing *de novo* femoral atherosclerosis over 6 years, but the presence or severity of atherosclerosis did not influence clonal expansion over the same period. The causal effect of CH is supported by several animal studies, particularly those investigating *TET2* mutations,[312] but has not yet been tested in clinical perturbation studies.

Therefore, while CH is inherently linked with aging, its association with age-related CVD suggests that it could both contribute to and result from systemic aging processes.

4.4 Imaging techniques

An arsenal of imaging techniques is available to help characterize cardiovascular aging. Echocardiography is the most widely used and accessible test of ventricular function and is vital in the diagnosis of HF with both reduced and preserved ejection fraction (HFrEF and HFpEF, respectively). HFpEF, especially, is largely a disease of aging, with ventricular stiffness in diastole commonly identified in older echo subjects.[313] Measures of diastolic dysfunction, such as a dilated left atrium (LA), reduced E/A ratio and increased E/e' ratio are useful measures of cardiovascular aging even in the absence of a diagnosis of HFpEF.[314] Atria abnormalities such as dilatation may represent 'atrial cardiopathy',[315] which is a strong predictor of both incident atrial fibrillation (AF) and stroke. However, clinical trials in patients with atrial cardiopathy, but not AF, have not yet yielded strong evidence for anticoagulant use.[316]

Measured with B-mode ultrasound, the carotid artery intima-media thickness (CIMT) is a measure of subclinical atherosclerosis and is well studied as a risk-stratifying tool.[317] Although a higher CIMT correlates with higher event rates, it remains unclear whether CIMT measurement in clinical practice improves outcomes, and it is therefore not recommended for this purpose in international guidelines.[318] CIMT increases robustly with age, and this appears to influence its predictive power, with one retrospective study finding CIMT improved risk stratification for cardiovascular death only in younger patients.[319] Each standard deviation increase in CIMT was associated with a 27% increased risk of cardiovascular death in the 35–44 years age group, but only a 14% increase in the 65–74 years group.[319] CIMT may therefore be most valuable as an assessment of premature cardiovascular aging, losing value in those who are already old.

Coronary calcium deposition is a hallmark of atherosclerosis and cardiovascular aging,[320,321] and CT imaging provides rapid, non-invasive estimation of the burden of calcium in coronary arteries—a CAC score.[322] CAC increases with age, and can therefore be used to give an 'estimated coronary age'.[323] This gives vital information in assessing risk for atherosclerotic CVD but also gives a powerful way to relate this risk to a patient.

In comparison to CT and ultrasonography, cardiac MRI has higher spatial resolution and so produces the most detailed assessment of myocardial tissue structure,[324,325] making it an attractive modality for multiparametric assessment of myocardial aging. Several studies have applied deep learning algorithms to the UK Biobank population, analysing genetic associations with markers of ventricular stiffness, diastolic dysfunction and aortic distensibility, among other MR evidence of aging.[326–328] These studies have shown that aortic and left atrial dimensions appear particularly predictive of aging and contribute the most to a synthesized age-prediction model based on imaging parameters.

Together, these varied imaging techniques allow powerful assessment of myocardial aging and risk stratification in older individuals.

4.5 Vascular function assessment

Arterial stiffness and endothelial dysfunction are a fundamental mechanism of many CVDs of aging, including hypertension, atherosclerosis and thrombosis[329] and both can be measured non-invasively.

The carotid-femoral pulse wave velocity (PVW) is the gold-standard non-invasive assessment of central arterial stiffness, with relatively simple and reproducible measurement and a large body of evidence supporting its association with CVD.[330–335] PVW uses electrocardiography and tonometry to measure the delay for an arterial pulsation to arrive at the carotid and femoral arteries, with stiffer arteries transmitting the pulsation more quickly. Notably, PVW values increase substantially with age, even in patients without CVD and normal blood pressure, and increase even more sharply in older patients with hypertension.[330] This suggests that arterial stiffness, as measured by PVW, is a usable surrogate marker for cardiovascular age.[336]

Coronary artery endothelial dysfunction in atherosclerosis was first demonstrated with paradoxical vasodilatation after injecting acetylcholine into diseases coronary arteries,[337] but technique is invasive and difficult to perform. Non-invasive assessments of endothelial dysfunction have since been introduced, including flow-mediated dilatation (FMD), which broadly assess the microvasculature, and peripheral arterial tonometry (PAT), assessing the microvasculature.[338]

FMD, which measures endothelium-dependent vasodilation, involves visualizing the brachial artery with ultrasound and then occluding the artery distally, using a cuff inflated to supra-systolic pressure. The dilatation of the brachial artery is then measured using ultrasound, with less dilatation indicating endothelial dysfunction. Lower FMD is associated with the presence and progression of atherosclerosis,[339,340] and the occurrence of cardiovascular events,[341–343] but its lack of incremental predictive power over traditional risk factors has prevented its recommendation in guidelines.[338]

An alternative measure of endothelial dysfunction in the PAT technique, in which a finger probe measures changes in the pulse waveform before and after reactive hyperaemia, which is induced with temporary occlusion of arterial flow with an inflated cuff.[344] This has the advantage of being much less operator-dependent than FMD measurement, although the body of evidence linking PAT measurements to CVD, while present, is less robust.[345,346]

5. Modelling cardiovascular ageing

Traditionally, statistical models have played a central role in estimating cardiovascular risk and the progression of cardiovascular aging. Among the most established models is the Framingham Risk Score, which estimates a 10-year risk of cardiovascular events based on key clinical variables such as age, blood pressure, cholesterol levels, smoking status, and the presence of diabetes.[347] Additional risk models like SCORE (Systematic COronary Risk Evaluation)[347,348] and QRISK[349] have been developed to tailor cardiovascular risk prediction to specific population groups. These traditional models predominantly use linear and logistic regression techniques, which assume a relatively linear progression of cardiovascular aging and focus on a restricted set of modifiable and non-modifiable risk factors.

While instrumental in clinical risk stratification, these models present significant limitations. They often struggle to capture complex, non-linear interactions among diverse biological processes and are not designed to integrate high-dimensional data sources, such as genomics, metabolomics, or imaging modalities. To address these shortcomings, systems biology approaches have emerged, leveraging the integration of multi-omics data—including genomic, transcriptomic, and proteomic profiles—alongside clinical and environmental variables to build more comprehensive predictive models of cardiovascular aging.[350] Network-based modelling further facilitates the identification of molecular pathways and biomarkers that

Table 1 Blood-based biological clocks from large or midsized studies that predict mortality and/or cardiovascular relevant endpoints (adapted from Liberale *et al.*, 2025)[148]

Blood-based biological clocks	Description	Predictive value for lifespan and/or healthspan
Haematological aging clock[355]	Several deep learning-based biological age predictors trained on 20 population-specific blood biomarkers and cell counts.	Associated with all-cause mortality
DNAmAge[354]	DNAm age is a molecular readout reflecting intrinsic aging processes and is defined as the predicted biological age	Associated with increased risk for all-cause mortality and CV disease. Diet and lifestyle treatment leads to a decrease in DNAmAge.
GlycanAge[356]	A biological age test measuring chronic inflammation via blood-based glycan profiles on IgG antibodies, which correlate with chronological age	Associated with multiple diseases, among others CV disease and diabetes.
PhenoAge[356]	An epigenetic clock based on DNA methylation at CpG sites strongly correlated with chronological age	Associated with risk of cancer, Alzheimer's disease, CHD.
Proteomic clocks[357]	Proteomic clocks use protein biomarkers as intermediate phenotypes closely linked to age-related diseases, offering potentially greater accuracy in assessing aging and pathology	May predict CV death
Metabolomic clock[358]	Metabolomic clocks assess metabolites and small molecules as key links between genotype and phenotype in aging and age-related disease	Associated with all-cause, CV, cancer- and infection-related mortality
iAGE (Inflammatory aging clock)[359]	A blood-based immune biomarker metric for chronic inflammation, used to predict aging phenotypes and understand vascular aging mechanisms	Associated with exceptional longevity in centenarians. Associated with multimorbidity, immunosenescence, frailty and CV aging.
IMM-AGE (Immune aging score)[360]	A high-dimensional immune aging trajectory that more accurately reflects immune status than chronological age.	Better performance in predicting mortality in older adults than the epigenetic clock.

underpin cardiovascular aging, providing a foundation for targeted therapeutic interventions.[351]

Recent findings underscore the utility of such integrative approaches. A study involving over 6200 middle-aged individuals demonstrated that organ-specific proteomic signatures are predictive of long-term risk for age-related diseases.[352] Notably, the study found high organ specificity for chronic HF and dilated cardiomyopathy among participants exhibiting a pronounced heart-age gap.

In parallel, advances in machine learning (ML) and artificial intelligence (AI) have revolutionized the modelling of cardiovascular aging. In contrast to traditional regression-based methods, ML techniques can manage extensive, multidimensional datasets and detect complex non-linear relationships that influence cardiovascular aging. Various ML strategies—including ensemble models that integrate random forests, support vector machines, neural networks, and deep learning—have been successfully applied to large datasets such as the UK Biobank, comprising over 375 000 individuals, to enhance cardiovascular risk prediction.[353] Notably, the incorporation of mental health questionnaire data into the ensemble algorithm significantly improved predictive accuracy, increasing CVD risk prediction from 71% to 85%.

Biological aging clocks, which estimate biological age and highlight the divergence from chronological age, may also be valuable tools in modelling cardiovascular aging and capturing individual aging dynamics.[354] An effective clock would also be sensitive to interventions or drug effects. Current clocks are based on measures such as epigenetic changes, inflammatory markers, plasma proteomics, or metabolomics (*Table 1*). However, their low intercorrelation suggests they capture distinct facets of the aging process.

In conclusion, the modelling of cardiovascular aging has progressed from conventional statistical models to sophisticated, AI-driven approaches and systems biology frameworks that offer deeper insights and improved predictive capability.

6. Future directions

The modelling of cardiovascular aging has progressed from conventional statistical models to AI-driven approaches and systems biology frameworks that offer deeper insights and improved predictive capability. By integrating multi-dimensional biomarkers, such as epigenetic clocks, proteomic profiles, and arterial stiffness measures, personalized models that can quantify biological age and capture individualized vascular aging trajectories will be a transformative frontier in preventive and precision medicine.[352,361,362] Furthermore, continuous monitoring allowing dynamic risk assessment through wearable technology and digital health tools can enhance these models by providing real-time data on physical and chemical signals that reflect the health conditions of older adults.[363] Sex-specific factors, including differential vascular biology, plaque vulnerability, and hormonal influences, must be systematically incorporated in these models to enhance predictive accuracy, particularly given the accelerated vascular stiffening observed in postmenopausal women and distinct CVD manifestations between sexes.[364–366] Emerging insights into epigenetic, clonal haematopoiesis and neuro-cardiovascular axes further underscore the need to expand biomarker panels to better reflect systemic aging processes.[301,367,368] In that view, Aβ40 holds great potential for predicting adverse outcomes across life.[141,142] However, critical challenges persist, including the standardization of measurements, validation of biomarkers across diverse ethnic, socioeconomic, and gender-diverse populations, and the establishment of causal links through large longitudinal studies. Ethical and regulatory frameworks must also evolve to support the translation of predictive models into clinical practice, particularly for preventive gerotherapeutics. By bridging mechanistic insights with clinical innovation, personalized models hold promise not only for mitigating CVD burden but also for redefining healthy aging paradigms in our increasingly diverse and aging global population.

Funding

L.S. is supported by a British Heart Foundation Clinical Research Training Fellowship (FS/CRTF/23/24520). J.H. is supported by a German Research Foundation/Deutsche Forschungsgemeinschaft (DFG) HA 2868/14-1. J.A. is supported by a German Research Foundation/Deutsche Forschungsgemeinschaft (DFG) AL288/5-1. S.T.-C. is supported by grants from the British Heart Foundation grant (PG/23/11093) and the Royal

Society (RG\R1\241197). K.S. is supported by the German Research Foundation Deutsche Forschungsgemeinschaft (DFG) (CRC1366 C07, project no. 394046768), the Health + Life Science Alliance Heidelberg Mannheim GmbH and the Helmholtz Institute for Translational AngioCardioScience (HI-TAC).

Conflict of interest: I.S. is supported by Kancera and Astra Zeneca with research grants.

References

1. Jiang M, Tian S, Liu S, Wang Y, Guo X, Huang T, Lin X, Belsky DW, Baccarelli AA, Gao X. Author correction: accelerated biological aging elevates the risk of cardiometabolic multimorbidity and mortality. *Nat Cardiovasc Res* 2024;**3**:883.
2. Justice J, Miller JD, Newman JC, Hashmi SK, Halter J, Austad SN, Barzilai N, Kirkland JL. Frameworks for proof-of-concept clinical trials of interventions that target fundamental aging processes. *J Gerontol A Biol Sci Med Sci* 2016;**71**:1415–1423.
3. Abdellatif M, Rainer PP, Sedej S, Kroemer G. Hallmarks of cardiovascular ageing. *Nat Rev Cardiol* 2023;**20**:754–777.
4. Liberale L, Camici GG. The role of vascular aging in atherosclerotic plaque development and vulnerability. *Curr Pharm Des* 2019;**25**:3098–3111.
5. Chirinos JA, Segers P, Hughes T, Townsend R. Large-artery stiffness in health and disease: JACC State-of-the-Art Review. *J Am Coll Cardiol* 2019;**74**:1237–1263.
6. Lim CJ, Cech TR. Shaping human telomeres: from shelterin and CST complexes to telomeric chromatin organization. *Nat Rev Mol Cell Biol* 2021;**22**:283–298.
7. Olovnikov AM. A theory of marginotomy. The incomplete copying of template margin in enzymic synthesis of polynucleotides and biological significance of the phenomenon. *J Theor Biol* 1973;**41**:181–190.
8. Di Micco R, Krizhanovsky V, Baker D, d'Adda di Fagagna F. Cellular senescence in ageing: from mechanisms to therapeutic opportunities. *Nat Rev Mol Cell Biol* 2021;**22**:75–95.
9. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. Hallmarks of aging: an expanding universe. *Cell* 2023;**186**:243–278.
10. Collins K. The biogenesis and regulation of telomerase holoenzymes. *Nat Rev Mol Cell Biol* 2006;**7**:484–494.
11. Yu HJ, Byun YH, Park CK. Techniques for assessing telomere length: a methodological review. *Comput Struct Biotechnol J* 2024;**23**:1489–1498.
12. Su Y, Yin L, Zhao Y, Zhao Y, Zhang W, Ke Y, Wang M, He X, Liu M, Liu G, Qin P, Hu F, Zhang M, Hu D. The association of telomere length and coronary heart disease: a systematic review and dose-response meta-analysis. *Nutr Metab Cardiovasc Dis* 2025;**35**:103830.
13. Aung N, Wang Q, van Duijvenboden S, Burns R, Stoma S, Raisi-Estabragh Z, Ahmet S, Allara E, Wood A, Di Angelantonio E, Danesh J, Munroe PB, Young A, Harvey NC, Codd V, Nelson CP, Petersen SE, Samani NJ. Association of longer leukocyte telomere length with cardiac size, function, and heart failure. *JAMA Cardiol* 2023;**8**:808–815.
14. van der Harst P, van der Steege G, de Boer RA, Voors AA, Hall AS, Mulder MJ, van Gilst WH, van Veldhuisen DJ; MERIT-HF Study Group. Telomere length of circulating leukocytes is decreased in patients with chronic heart failure. *J Am Coll Cardiol* 2007;**49**:1459–1464.
15. Spyridopoulos I, Hoffmann J, Aicher A, Brümmendorf TH, Doerr HW, Zeiher AM, Dimmeler S. Accelerated telomere shortening in leukocyte subpopulations of patients with coronary heart disease: role of cytomegalovirus seropositivity. *Circulation* 2009;**120**:1364–1372.
16. Spyridopoulos I, Erben Y, Brummendorf TH, Haendeler J, Dietz K, Seeger F, Kissel CK, Martin H, Hoffmann J, Assmus B, Zeiher AM, Dimmeler S. Telomere gap between granulocytes and lymphocytes is a determinant for hematopoetic progenitor cell impairment in patients with previous myocardial infarction. *Arterioscler Thromb Vasc Biol* 2008;**28**:968–974.
17. Smith GD. Mendelian randomization for strengthening causal inference in observational studies: application to gene × environment interactions. *Perspect Psychol Sci* 2010;**5**:527–545.
18. Li W, Liu C, Chen Y, Dong S, Zhang M, Sun J, Zhao Z, Zuo Y, Chen S. Assessment of the relationship between telomere length and atherosclerosis: a Mendelian randomization study. *Medicine (Baltimore)* 2023;**102**:e35875.
19. Scheller Madrid A, Rode L, Nordestgaard BG, Bojesen SE. Short telomere length and ischemic heart disease: observational and genetic studies in 290 022 individuals. *Clin Chem* 2016;**62**:1140–1149.
20. Chung HG, Yang PS, Cho S, Jang E, Kim D, Yu HT, Kim TH, Uhm JS, Sung JH, Pak HN, Lee MH, Joung B. The associations of leukocyte telomere length and intermediary cardiovascular phenotype with adverse cardiovascular outcomes in the white population. *Sci Rep* 2024;**14**:13975.
21. Codd V, Wang Q, Allara E, Musicha C, Kaptoge S, Stoma S, Jiang T, Hamby SE, Braund PS, Bountziouka V, Budgeon CA, Denniff M, Swinfield C, Papakonstantinou M, Sheth S, Nanus DE, Warner SC, Wang M, Khera AV, Eales J, Ouwehand WH, Thompson JR, Di Angelantonio E, Wood AM, Butterworth AS, Danesh JN, Nelson CP, Samani NJ. Polygenic basis and biomedical consequences of telomere length variation. *Nat Genet* 2021;**53**:1425–1433.
22. Li B, Xiong W, Zuo W, Shi Y, Wang T, Chang L, Wu Y, Ma H, Bian Q, Chang ACY. Proximal telomeric decompaction due to telomere shortening drives FOXC1-dependent myocardial senescence. *Nucleic Acids Res* 2024;**52**:6269–6284.
23. Spinelli JB, Haigis MC. The multifaceted contributions of mitochondria to cellular metabolism. *Nat Cell Biol* 2018;**20**:745–754.
24. Tan JX, Finkel T. Mitochondria as intracellular signaling platforms in health and disease. *J Cell Biol* 2020;**219**:e202002179.
25. Popov LD. Mitochondria as intracellular signalling organelles. An update. *Cell Signal* 2023;**109**:110794.
26. Mone P, Agyapong ED, Morciano G, Jankauskas SS, De Luca A, Varzideh F, Pinton P, Santulli G. Dysfunctional mitochondria elicit bioenergetic decline in the aged heart. *J Cardiovasc Aging* 2024;**4**:13.
27. Iakovou E, Kourti M. A comprehensive overview of the complex role of oxidative stress in aging, the contributing environmental stressors and emerging antioxidant therapeutic interventions. *Front Aging Neurosci* 2022;**14**:827900.
28. Giorgi C, Marchi S, Simoes ICM, Ren Z, Morciano G, Perrone M, Patalas-Krawczyk P, Borchard S, Jędrak P, Pierzynowska K, Szymański J, Wang DQ, Portincasa P, Węgrzyn G, Zischka H, Dobrzyn P, Bonora M, Duszynski J, Rimessi A, Karkucinska-Wieckowska A, Dobrzyn A, Szabadkai G, Zavan B, Oliveira PJ, Sardao VA, Pinton P, Wieckowski MR. Mitochondria and reactive oxygen Species in aging and age-related diseases. *Int Rev Cell Mol Biol* 2018;**340**:209–344.
29. Qu K, Yan F, Qin X, Zhang K, He W, Dong M, Wu G. Mitochondrial dysfunction in vascular endothelial cells and its role in atherosclerosis. *Front Physiol* 2022;**13**:1084604.
30. Xia Y, Zhang X, An P, Luo J, Luo Y. Mitochondrial homeostasis in VSMCs as a central hub in vascular remodeling. *Int J Mol Sci* 2023;**24**:3483.
31. Li X, Zhang W, Cao Q, Wang Z, Zhao M, Xu L, Zhuang Q. Mitochondrial dysfunction in fibrotic diseases. *Cell Death Discov* 2020;**6**:80.
32. Yang J, Guo Q, Feng X, Liu Y, Zhou Y. Mitochondrial dysfunction in cardiovascular diseases: potential targets for treatment. *Front Cell Dev Biol* 2022;**10**:841523.
33. Greider CW, Blackburn EH. Identification of a specific telomere terminal transferase activity in Tetrahymena extracts. *Cell* 1985;**43**:405–413.
34. Morin GB. The human telomere terminal transferase enzyme is a ribonucleoprotein that synthesizes TTAGGG repeats. *Cell* 1989;**59**:521–529.
35. Santos JH, Meyer JN, Skorvaga M, Annab LA, Van Houten B. Mitochondrial hTERT exacerbates free-radical-mediated mtDNA damage. *Aging Cell* 2004;**3**:399–411.
36. Ahmed S, Passos JF, Birket MJ, Beckmann T, Brings S, Peters H, Birch-Machin MA, von Zglinicki T, Saretzki G. Telomerase does not counteract telomere shortening but protects mitochondrial function under oxidative stress. *J Cell Sci* 2008;**121**:1046–1053.
37. Haendeler J, Dröse S, Büchner N, Jakob S, Altschmied J, Goy C, Spyridopoulos I, Zeiher AM, Brandt U, Dimmeler S. Mitochondrial telomerase reverse transcriptase binds to and protects mitochondrial DNA and function from damage. *Arterioscler Thromb Vasc Biol* 2009;**29**:929–935.
38. Spilsbury A, Miwa S, Attems J, Saretzki G. The role of telomerase protein TERT in Alzheimer's disease and in tau-related pathology in vitro. *J Neurosci* 2015;**35**:1659–1674.
39. Chung J, Khadka P, Chung IK. Nuclear import of hTERT requires a bipartite nuclear localization signal and Akt-mediated phosphorylation. *J Cell Sci* 2012;**125**:2684–2697.
40. Kovalenko OA, Caron MJ, Ulema P, Medrano C, Thomas AP, Kimura M, Bonini MG, Herbig U, Santos JH. A mutant telomerase defective in nuclear-cytoplasmic shuttling fails to immortalize cells and is associated with mitochondrial dysfunction. *Aging Cell* 2010;**9**:203–219.
41. Sharma NK, Reyes A, Green P, Caron MJ, Bonini MG, Gordon DM, Holt IJ, Santos JH. Human telomerase acts as a hTR-independent reverse transcriptase in mitochondria. *Nucleic Acids Res* 2012;**40**:712–725.
42. Ait-Aissa K, Heisner JS, Norwood Toro LE, Bruemmer D, Doyon G, Harmann L, Geurts A, Camara AKS, Beyer AM. Telomerase deficiency predisposes to heart failure and ischemia-reperfusion injury. *Front Cardiovasc Med* 2019;**6**:31.
43. Ait-Aissa K, Norwood-Toro LE, Terwoord J, Young M, Paniagua LA, Hader SN, Hughes WE, Hockenberry JC, Beare JE, Linn J, Kohmoto T, Kim J, Betts DH, LeBlanc AJ, Gutterman DD, Beyer AM. Noncanonical role of telomerase in regulation of microvascular redox environment with implications for coronary artery disease. *Function (Oxf)* 2022;**3**:zqac043.
44. Ale-Agha N, Jakobs P, Goy C, Zurek M, Rosen J, Dyballa-Rukes N, Metzger S, Greulich J, von Ameln F, Eckermann O, Unfried K, Brack F, Grandoch M, Thielmann M, Kamler M, Gedik N, Kleinbongard P, Heinen A, Heusch G, Gödecke A, Altschmied J, Haendeler J. Mitochondrial telomerase reverse transcriptase protects from myocardial ischemia/reperfusion injury by improving Complex I composition and function. *Circulation* 2021;**144**:1876–1890.
45. Wan T, Weir EJ, Johnson M, Korolchuk VI, Saretzki GC. Increased telomerase improves motor function and alpha-synuclein pathology in a transgenic mouse model of Parkinson's disease associated with enhanced autophagy. *Prog Neurobiol* 2021;**199**:101953.
46. Miwa S, Czapiewski R, Wan T, Bell A, Hill KN, von Zglinicki T, Saretzki G. Decreased mTOR signalling reduces mitochondrial ROS in brain via accumulation of the telomerase protein TERT within mitochondria. *Aging (Albany NY)* 2016;**8**:2551–2567.
47. Kleinbongard P, Gedik N, Kirca M, Stoian L, Frey U, Zandi A, Thielmann M, Jakob H, Peters J, Kamler M, Heusch G. Mitochondrial and contractile function of human right atrial tissue in response to remote ischemic conditioning. *J Am Heart Assoc* 2018;**7**:e009540.
48. Hernandez-Segura A, Nehme J, Demaria M. Hallmarks of cellular senescence. *Trends Cell Biol* 2018;**28**:436–453.
49. Childs BG, Durik M, Baker DJ, van Deursen JM. Cellular senescence in aging and age-related disease: from mechanisms to therapy. *Nat Med* 2015;**21**:1424–1435.

50. Hayflick L, Moorhead PS. The serial cultivation of human diploid cell strains. *Exp Cell Res* 1961;**25**:585–621.
51. Minamino T, Miyauchi H, Yoshida T, Ishida Y, Yoshida H, Komuro I. Endothelial cell senescence in human atherosclerosis. *Circulation* 2002;**105**:1541–1544.
52. Wang J, Uryga AK, Reinhold J, Figg N, Baker L, Finigan A, Gray K, Kumar S, Clarke M, Bennett M. Vascular smooth muscle cell senescence promotes atherosclerosis and features of plaque vulnerability. *Circulation* 2015;**132**:1909–1919.
53. Dookun E, Walaszczyk A, Redgrave R, Palmowski P, Tual-Chalot S, Suwana A, Chapman J, Jirkovsky E, Donastorg Sosa L, Gill E, Yausep OE, Santin Y, Mialet-Perez J, Andrew Owens W, Grieve D, Spyridopoulos I, Taggart M, Arthur HM, Passos JF, Richardson GD. Clearance of senescent cells during cardiac ischemia-reperfusion injury improves recovery. *Aging Cell* 2020;**19**:e13249.
54. Katoh M, Nomura S, Yamada S, Ito M, Hayashi H, Katagiri M, Heryed T, Fujiwara T, Takeda N, Nishida M, Sugaya M, Kato M, Osawa T, Abe H, Sakurai Y, Ko T, Fujita K, Zhang B, Hatsuse S, Yamada T, Inoue S, Dai Z, Kubota M, Sawami K, Ono M, Morita H, Kubota Y, Mizuno S, Takahashi S, Nakanishi M, Ushiku T, Nakagami H, Aburatani H, Komuro I. Vaccine therapy for heart failure targeting the inflammatory cytokine Igfbp7. *Circulation* 2024;**150**:374–389.
55. Booth LK, Redgrave RE, Tual-Chalot S, Spyridopoulos I, Phillips HM, Richardson GD. Heart disease and ageing: the roles of senescence, mitochondria, and telomerase in cardiovascular disease. *Subcell Biochem* 2023;**103**:45–78.
56. Chen JH, Hales CN, Ozanne SE. DNA damage, cellular senescence and organismal ageing: causal or correlative? *Nucleic Acids Res* 2007;**35**:7417–7428.
57. Narasimhan A, Flores RR, Robbins PD, Niedernhofer LJ. Role of cellular senescence in type II diabetes. *Endocrinology* 2021;**162**:bqab136.
58. Hauck AK, Bernlohr DA. Oxidative stress and lipotoxicity. *J Lipid Res* 2016;**57**:1976–1986.
59. Smith U, Li Q, Rydén M, Spalding KL. Cellular senescence and its role in white adipose tissue. *Int J Obes* 2021;**45**:934–943.
60. Tchkonia T, Morbeck DE, Von Zglinicki T, Van Deursen J, Lustgarten J, Scrable H, Khosla S, Jensen MD, Kirkland JL. Fat tissue, aging, and cellular senescence. *Aging Cell* 2010;**9**:667–684.
61. Minamino T, Orimo M, Shimizu I, Kunieda T, Yokoyama M, Ito T, Nojima A, Nabetani A, Oike Y, Matsubara H, Ishikawa F, Komuro I. A crucial role for adipose tissue p53 in the regulation of insulin resistance. *Nat Med* 2009;**15**:1082–1087.
62. Chen YW, Harris RA, Hatahet Z, Chou KM. Ablation of XP-V gene causes adipose tissue senescence and metabolic abnormalities. *Proc Natl Acad Sci U S A* 2015;**112**:E4556–E4564.
63. Vergoni B, Cornejo PJ, Gilleron J, Djedaini M, Ceppo F, Jacquel A, Bouget G, Ginet C, Gonzalez T, Maillet J, Dhennin V, Verbanck M, Auberger P, Froguel P, Tanti JF, Cormont M. DNA damage and the activation of the p53 pathway mediate alterations in metabolic and secretory functions of adipocytes. *Diabetes* 2016;**65**:3062–3074.
64. Nyunoya T, Monick MM, Klingelhutz A, Yarovinsky TO, Cagley JR, Hunninghake GW. Cigarette smoke induces cellular senescence. *Am J Respir Cell Mol Biol* 2006;**35**:681–688.
65. Anagnostou D, Theodorakis N, Hitas C, Kreouzi M, Pantos I, Vamvakou G, Nikolaou M. Sarcopenia and cardiogeriatrics: the links between Skeletal muscle decline and cardiovascular aging. *Nutrients* 2025;**17**:282.
66. Korolchuk VI, Miwa S, Carroll B, von Zglinicki T. Mitochondria in cell senescence: is mitophagy the weakest link? *EBioMedicine* 2017;**21**:7–13.
67. Kelly G, Kataura T, Panek J, Ma G, Salmonowicz H, Davis A, Kendall H, Brookes C, Ayine-Tora DM, Banks P, Nelson G, Dobby L, Pitrez PR, Booth L, Costello L, Richardson GD, Lovat P, Przyborski S, Ferreira L, Greaves L, Szczepanowska K, von Zglinicki T, Miwa S, Brown M, Flagler M, Oblong JE, Bascom CC, Carroll B, Reynisson J, Korolchuk VI. Suppressed basal mitophagy drives cellular aging phenotypes that can be reversed by a p62-targeting small molecule. *Dev Cell* 2024;**59**:1924–1939.e7.
68. Taneike M, Yamaguchi O, Nakai A, Hikoso S, Takeda T, Mizote I, Oka T, Tamai T, Oyabu J, Murakawa T, Nishida K, Shimizu T, Hori M, Komuro I, Takuji Shirasawa TS, Mizushima N, Otsu K. Inhibition of autophagy in the heart induces age-related cardiomyopathy. *Autophagy* 2010;**6**:600–606.
69. Belenchia AM, Gavini MP, Toedebusch RG, DeMarco VG, Pulakat L. Comparison of cardiac miRNA transcriptomes induced by diabetes and Rapamycin treatment and identification of a rapamycin-associated cardiac MicroRNA signature. *Oxid Med Cell Longev* 2018;**2018**:8364608.
70. Pulakat L, Chen HH. Pro-senescence and anti-senescence mechanisms of cardiovascular aging: cardiac MicroRNA regulation of longevity drug-induced autophagy. *Front Pharmacol* 2020;**11**:774.
71. Tyrrell DJ, Blin MG, Song J, Wood SC, Zhang M, Beard DA, Goldstein DR. Age-associated mitochondrial dysfunction accelerates atherogenesis. *Circ Res* 2020;**126**:298–314.
72. Correia-Melo C, Passos JF. Mitochondria: are they causal players in cellular senescence? *Biochim Biophys Acta* 2015;**1847**:1373–1379.
73. Ding W, Chen J, Zhao L, Wu S, Chen X, Chen H. Mitochondrial DNA leakage triggers inflammation in age-related cardiovascular diseases. *Front Cell Dev Biol* 2024;**12**:1287447.
74. Correia-Melo C, Marques FD, Anderson R, Hewitt G, Hewitt R, Cole J, Carroll BM, Miwa S, Birch J, Merz A, Rushton MD, Charles M, Jurk D, Tait SW, Czapiewski R, Greaves L, Nelson G, Bohlooly-Y M, Rodriguez-Cuenca S, Vidal-Puig A, Mann D, Saretzki G, Quarato G, Green DR, Adams PD, von Zglinicki T, Korolchuk VI, Passos JF. Mitochondria are required for pro-ageing features of the senescent phenotype. *EMBO J* 2016;**35**:724–742.
75. Birch J, Passos JF. Targeting the SASP to combat ageing: mitochondria as possible intracellular allies? *Bioessays* 2017;**39**:1–7.
76. Civril F, Deimling T, de Oliveira Mann CC, Ablasser A, Moldt M, Witte G, Hornung V, Hopfner KP. Structural mechanism of cytosolic DNA sensing by cGAS. *Nature* 2013;**498**:332–337.
77. Vizioli MG, Liu T, Miller KN, Robertson NA, Gilroy K, Lagnado AB, Perez-Garcia A, Kiourtis C, Dasgupta N, Lei X, Kruger PJ, Nixon C, Clark W, Jurk D, Bird TG, Passos JF, Berger SL, Dou Z, Adams PD. Mitochondria-to-nucleus retrograde signaling drives formation of cytoplasmic chromatin and inflammation in senescence. *Genes Dev* 2020;**34**:428–445.
78. Eirin A, Herrmann SM, Saad A, Abumoawad A, Tang H, Lerman A, Textor SC, Lerman LO. Urinary mitochondrial DNA copy number identifies renal mitochondrial injury in renovascular hypertensive patients undergoing renal revascularization: a pilot study. *Acta Physiol (Oxf)* 2019;**226**:e13267.
79. Cosentino N, Campodonico J, Moltrasio M, Lucci C, Milazzo V, Rubino M, De Metrio M, Marana I, Grazi M, Bonomi A, Veglia F, Lauri G, Bartorelli AL, Marenzi G. Mitochondrial biomarkers in patients with ST-elevation myocardial infarction and their potential prognostic implications: a prospective observational study. *J Clin Med* 2021;**10**:275.
80. Decout A, Katz JD, Venkatraman S, Ablasser A. The cGAS-STING pathway as a therapeutic target in inflammatory diseases. *Nat Rev Immunol* 2021;**21**:548–569.
81. Walaszczyk A, Dookun E, Redgrave R, Tual-Chalot S, Victorelli S, Spyridopoulos I, Owens A, Arthur HM, Passos JF, Richardson GD. Pharmacological clearance of senescent cells improves survival and recovery in aged mice following acute myocardial infarction. *Aging Cell* 2019;**18**:e12945.
82. Algoet M, Janssens S, Himmelreich U, Gsell W, Pusovnik M, Van den Eynde J, Oosterlinck W. Myocardial ischemia-reperfusion injury and the influence of inflammation. *Trends Cardiovasc Med* 2023;**33**:357–366.
83. Lewis-McDougall FC, Ruchaya PJ, Domenjo-Vila E, Shin Teoh T, Prata L, Cottle BJ, Clark JE, Punjabi PP, Awad W, Torella D, Tchkonia T, Kirkland JL, Ellison-Hughes GM. Aged-senescent cells contribute to impaired heart regeneration. *Aging Cell* 2019;**18**:e12931.
84. Linders AN, Dias IB, Ovchinnikova ES, Vermeer MCSC, Hoes MF, Markousis Mavrogenis G, Deiman FE, Arevalo Gomez KF, Bliley JM, Nehme J, Vink A, Gietema J, de Boer RA, Westenbrink D, Sillje HHW, Hilfiker-Kleiner D, van Laake LW, Feinberg AW, Demaria M, Bomer N, van der Meer P. Evaluation of senescence and its prevention in doxorubicin-induced cardiotoxicity using dynamic engineered heart tissues. *JACC: Cardio Oncology* 2023;**5**:298–315.
85. Booth LN, Brunet A. The aging epigenome. *Mol Cell* 2016;**62**:728–744.
86. Rios FJ, de Ciuceis C, Georgiopoulos G, Lazaridis A, Nosalski R, Pavlidis G, Tual-Chalot S, Agabiti-Rosei C, Camargo LL, Dąbrowska E, Quarti-Trevano F, Hellmann M, Masi S, Lopreiato M, Mavraganis G, Mengozzi A, Montezano AC, Stavropoulos K, Winklewski PJ, Wolf J, Costantino S, Doumas M, Gkaliagkousi E, Grassi G, Guzik TJ, Ikonomidis I, Narkiewicz K, Paneni F, Rizzoni D, Stamatelopoulos K, Stellos K, Taddei S, Touyz RM, Virdis A. Mechanisms of vascular inflammation and potential therapeutic targets: a position paper from the ESH Working Group on small arteries. *Hypertension* 2024;**81**:1218–1232.
87. Gems D, Virk RS, de Magalhaes JP. Epigenetic clocks and programmatic aging. *Ageing Res Rev* 2024;**101**:102546.
88. Sen P, Shah PP, Nativio R, Berger SL. Epigenetic mechanisms of longevity and aging. *Cell* 2016;**166**:822–839.
89. Yang JH, Hayano M, Griffin PT, Amorim JA, Bonkowski MS, Apostolides JK, Salfati EL, Blanchette M, Munding EM, Bhakta M, Chew YC, Guo W, Yang X, Maybury-Lewis S, Tian X, Ross JM, Coppotelli G, Meer MV, Rogers-Hammond R, Vera DL, Lu YR, Pippin JW, Creswell ML, Dou Z, Xu C, Mitchell SJ, Das A, O'Connell BL, Thakur S, Kane AE, Su Q, Mohri Y, Nishimura EK, Schaevitz L, Garg N, Balta AM, Rego MA, Gregory-Ksander M, Jakobs TC, Zhong L, Wakimoto H, El Andari J, Grimm D, Mostoslavsky R, Wagers AJ, Tsubota K, Bonasera SJ, Palmeira CM, Seidman JG, Seidman CE, Wolf NS, Kreiling JA, Sedivy JM, Murphy GF, Green RE, Garcia BA, Berger SL, Oberdoerffer P, Shankland SJ, Gladyshev VN, Ksander BR, Pfenning AR, Rajman LA, Sinclair DA. Loss of epigenetic information as a cause of mammalian aging. *Cell* 2023;**186**:305–326.e27.
90. Hall E, Volkov P, Dayeh T, Esguerra JL, Salö S, Eliasson L, Rönn T, Bacos K, Ling C. Sex differences in the genome-wide DNA methylation pattern and impact on gene expression, microRNA levels and insulin secretion in human pancreatic islets. *Genome Biol* 2014;**15**:522.
91. Asllanaj E, Zhang X, Ochoa Rosales C, Nano J, Bramer WM, Portilla-Fernandez E, Braun KVE, Gonzalez-Jaramillo V, Ahrens W, Ikram A, Ghanbari M, Voortman T, Franco OH, Muka T, Glisic M. Sexually dimorphic DNA-methylation in cardiometabolic health: a systematic review. *Maturitas* 2020;**135**:6–26.
92. Zhu X, Chen Z, Shen W, Huang G, Sedivy JM, Wang H, Ju Z. Inflammation, epigenetics, and metabolism converge to cell senescence and ageing: the regulation and intervention. *Signal Transduct Target Ther* 2021;**6**:245.
93. Sánchez-Cabo F, Fuster V, Silla-Castro JC, González G, Lorenzo-Vivas E, Alvarez R, Callejas S, Benguría A, Gil E, Núñez E, Oliva B, Mendiguren JM, Cortes-Canteli M, Bueno H, Andrés V, Ordovás JM, Fernández-Friera L, Quesada AJ, Garcia JM, Rossello X, Vázquez J, Dopazo A, Fernández-Ortiz A, Ibáñez B, Fuster JJ, Lara-Pezzi E. Subclinical atherosclerosis and accelerated epigenetic age mediated by inflammation: a multi-omics study. *Eur Heart J* 2023;**44**:2698–2709.
94. Khvorova A. Oligonucleotide therapeutics—a new class of cholesterol-lowering drugs. *N Engl J Med* 2017;**376**:4–7.
95. Yaskolka Meir A, Keller M, Müller L, Bernhart SH, Tsaban G, Zelicha H, Rinott E, Kaplan A, Gepner Y, Shelef I, Schwarzfuchs D, Ceglarek U, Stadler P, Blüher M, Stumvoll M, Kovacs P.

Shai I effects of lifestyle interventions on epigenetic signatures of liver fat: central randomized controlled trial. *Liver Int* 2021;**41**:2101–2111.
96. Maegawa S, Lu Y, Tahara T, Lee JT, Madzo J, Liang S, Jelinek J, Colman RJ, Issa JJ. Caloric restriction delays age-related methylation drift. *Nat Commun* 2017;**8**:539.
97. Schubeler D. Function and information content of DNA methylation. *Nature* 2015;**517**: 321–326.
98. Horvath S. DNA methylation age of human tissues and cell types. *Genome Biol* 2013;**14**: R115.
99. Jaiswal S, Libby P. Clonal haematopoiesis: connecting ageing and inflammation in cardiovascular disease. *Nat Rev Cardiol* 2020;**17**:137–144.
100. Fuster JJ, MacLauchlan S, Zuriaga MA, Polackal MN, Ostriker AC, Chakraborty R, Wu CL, Sano S, Muralidharan S, Rius C, Vuong J, Jacob S, Muralidhar V, Robertson AA, Cooper MA, Andrés V, Hirschi KK, Martin KA, Walsh K. Clonal hematopoiesis associated with TET2 deficiency accelerates atherosclerosis development in mice. *Science* 2017;**355**:842–847.
101. Sano S, Oshima K, Wang Y, Katanasaka Y, Sano M, Walsh K. CRISPR-mediated gene editing to assess the roles of Tet2 and Dnmt3a in clonal hematopoiesis and cardiovascular disease. *Circ Res* 2018;**123**:335–341.
102. Sano S, Oshima K, Wang Y, MacLauchlan S, Katanasaka Y, Sano M, Zuriaga MA, Yoshiyama M, Goukassian D, Cooper MA, Fuster JJ, Walsh K. Tet2-mediated clonal hematopoiesis accelerates heart failure through a mechanism involving the IL-1beta/NLRP3 inflammasome. *J Am Coll Cardiol* 2018;**71**:875–886.
103. Koch Z, Li A, Evans DS, Cummings S, Ideker T. Somatic mutation as an explanation for epigenetic aging. *Nat Aging* 2025;**5**:709–719.
104. Marttila S, Kananen L, Häyrynen S, Jylhävä J, Nevalainen T, Hervonen A, Jylhä M, Nykter M, Hurme M. Ageing-associated changes in the human DNA methylome: genomic locations and effects on gene expression. *BMC Genomics* 2015;**16**:179.
105. Jones MJ, Goodman SJ, Kobor MS. DNA methylation and healthy human aging. *Aging Cell* 2015;**14**:924–932.
106. Benayoun BA, Pollina EA, Brunet A. Epigenetic regulation of ageing: linking environmental inputs to genomic stability. *Nat Rev Mol Cell Biol* 2015;**16**:593–610.
107. Zampieri M, Ciccarone F, Calabrese R, Franceschi C, Burkle A, Caiafa P. Reconfiguration of DNA methylation in aging. *Mech Ageing Dev* 2015;**151**:60–70.
108. Zaina S, Heyn H, Carmona FJ, Varol N, Sayols S, Condom E, Ramírez-Ruz J, Gomez A, Gonçalves I, Moran S, Esteller M. DNA methylation map of human atherosclerosis. *Circ Cardiovasc Genet* 2014;**7**:692–700.
109. Yu J, Qiu Y, Yang J, Bian S, Chen G, Deng M, Kang H, Huang L. DNMT1-PPARgamma pathway in macrophages regulates chronic inflammation and atherosclerosis development in mice. *Sci Rep* 2016;**6**:30053.
110. Jiang YZ, Manduchi E, Stoeckert CJ Jr, Davies PF. Arterial endothelial methylome: differential DNA methylation in athero-susceptible disturbed flow regions in vivo. *BMC Genomics* 2015;**16**:506.
111. Jiang YZ, Jimenez JM, Ou K, McCormick ME, Zhang LD, Davies PF. Hemodynamic disturbed flow induces differential DNA methylation of endothelial Kruppel-like factor 4 promoter in vitro and in vivo. *Circ Res* 2014;**115**:32–43.
112. Sullivan KE, Reddy AB, Dietzmann K, Suriano AR, Kocieda VP, Stewart M, Bhatia M. Epigenetic regulation of tumor necrosis factor alpha. *Mol Cell Biol* 2007;**27**:5147–5160.
113. Bruunsgaard H, Skinhoj P, Pedersen AN, Schroll M, Pedersen BK. Ageing, tumour necrosis factor-alpha (TNF-alpha) and atherosclerosis. *Clin Exp Immunol* 2000;**121**:255–260.
114. Davizon-Castillo P, McMahon B, Aguila S, Bark D, Ashworth K, Allawzi A, Campbell RA, Montenont E, Nemkov T, D'Alessandro A, Clendenen N, Shih L, Sanders NA, Higa K, Cox A, Padilla-Romo Z, Hernandez G, Wartchow E, Trahan GD, Nozik-Grayck E, Jones K, Pietras EM, DeGregori J, Rondina MT, Di Paola J. TNF-alpha-driven inflammation and mitochondrial dysfunction define the platelet hyperreactivity of aging. *Blood* 2019;**134**: 727–740.
115. Matt SM, Lawson MA, Johnson RW. Aging and peripheral lipopolysaccharide can modulate epigenetic regulators and decrease IL-1beta promoter DNA methylation in microglia. *Neurobiol Aging* 2016;**47**:1–9.
116. Lian BSX, Kawasaki T, Kano N, Ori D, Ikegawa M, Isotani A, Kawai T. Regulation of Il6 expression by single CpG methylation in downstream of Il6 transcription initiation site. *iScience* 2022;**25**:104118.
117. Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, Fonseca F, Nicolau J, Koenig W, Anker SD, Kastelein JJP, Cornel JH, Pais P, Pella D, Genest J, Cifkova R, Lorenzatti A, Forster T, Kobalava Z, Vida-Simiti L, Flather M, Shimokawa H, Ogawa H, Dellborg M, Rossi PRF, Troquay RPT, Libby P, Glynn RJ; CANTOS Trial Group. Antiinflammatory therapy with canakinumab for atherosclerotic disease. *N Engl J Med* 2017;**377**:1119–1131.
118. Ridker PM, Libby P, MacFadyen JG, Thuren T, Ballantyne C, Fonseca F, Koenig W, Shimokawa H, Everett BM, Glynn RJ. Modulation of the interleukin-6 signalling pathway and incidence rates of atherosclerotic events and all-cause mortality: analyses from the Canakinumab Anti-Inflammatory Thrombosis Outcomes Study (CANTOS). *Eur Heart J* 2018;**39**:3499–3507.
119. Svensson EC, Madar A, Campbell CD, He Y, Sultan M, Healey ML, Xu H, D'Aco K, Fernandez A, Wache-Mainier C, Libby P, Ridker PM, Beste MT, Basson CT. TET2-Driven Clonal hematopoiesis and response to canakinumab: an exploratory analysis of the CANTOS randomized clinical trial. *JAMA Cardiol* 2022;**7**:521–528.
120. Kane AE, Sinclair DA. Epigenetic changes during aging and their reprogramming potential. *Crit Rev Biochem Mol Biol* 2019;**54**:61–83.
121. Cheung P, Vallania F, Warsinske HC, Donato M, Schaffert S, Chang SE, Dvorak M, Dekker CL, Davis MM, Utz PJ, Khatri P, Kuo AJ. Single-cell chromatin modification profiling reveals increased epigenetic variations with aging. *Cell* 2018;**173**:1385–1397.e14.
122. Dang W, Steffen KK, Perry R, Dorsey JA, Johnson FB, Shilatifard A, Kaeberlein M, Kennedy BK, Berger SL. Histone H4 lysine 16 acetylation regulates cellular lifespan. *Nature* 2009; **459**:802–807.
123. Narita M, Nũnez S, Heard E, Narita M, Lin AW, Hearn SA, Spector DL, Hannon GJ, Lowe SW. Rb-mediated heterochromatin formation and silencing of E2F target genes during cellular senescence. *Cell* 2003;**113**:703–716.
124. Cohn RL, Gasek NS, Kuchel GA, Xu M. The heterogeneity of cellular senescence: insights at the single-cell level. *Trends Cell Biol* 2023;**33**:9–17.
125. Wang K, Liu H, Hu Q, Wang L, Liu J, Zheng Z, Zhang W, Ren J, Zhu F, Liu GH. Epigenetic regulation of aging: implications for interventions of aging and diseases. *Signal Transduct Target Ther* 2022;**7**:374.
126. Sidler C, Kovalchuk O, Kovalchuk I. Epigenetic regulation of cellular senescence and aging. *Front Genet* 2017;**8**:138.
127. Mengozzi A, Costantino S, Paneni F, Duranti E, Nannipieri M, Mancini R, Lai M, La Rocca V, Puxeddu I, Antonioli L, Fornai M, Ghionzoli M, Georgiopoulos G, Ippolito C, Bernardini N, Ruschitzka F, Pugliese NR, Taddei S, Virdis A, Masi S. Targeting SIRT1 rescues age- and obesity-induced microvascular dysfunction in ex vivo human vessels. *Circ Res* 2022;**131**: 476–491.
128. Hu C, Peng K, Wu Q, Wang Y, Fan X, Zhang DM, Passerini AG, Sun C. HDAC1 and 2 regulate endothelial VCAM-1 expression and atherogenesis by suppressing methylation of the GATA6 promoter. *Theranostics* 2021;**11**:5605–5619.
129. Oksala NKJ, Seppälä I, Rahikainen R, Mäkelä KM, Raitoharju E, Illig T, Klopp N, Kholova I, Laaksonen R, Karhunen PJ, Hytönen VP, Lehtimäki T. Synergistic expression of histone deacetylase 9 and matrix metalloproteinase 12 in M4 macrophages in advanced carotid plaques. *Eur J Vasc Endovasc Surg* 2017;**53**:632–640.
130. CARDIoGRAMplusC4D Consortium; Deloukas P, Kanoni S, Willenborg C, Farrall M, Assimes TL, Thompson JR, Ingelsson E, Saleheen D, Erdmann J, Goldstein BA, Stirrups K, König IR, Cazier JB, Johansson A, Hall AS, Lee JY, Willer CJ, Chambers JC, Esko T, Folkersen L, Goel A, Grundberg E, Havulinna AS, Ho WK, Hopewell JC, Eriksson N, Kleber ME, Kristiansson K, Lundmark P, Lyytikäinen LP, Rafelt S, Shungin D, Strawbridge RJ, Thorleifsson G, Tikkanen E, Van Zuydam N, Voight BF, Waite LL, Zhang W, Ziegler A, Absher D, Altshuler D, Balmforth AJ, Barroso I, Braund PS, Burgdorf C, Claudi-Boehm S, Cox D, Dimitriou M, Do R; DIAGRAM Consortium; CARDIOGENICS Consortium; Doney AS, El Mokhtari N, Eriksson P, Fischer K, Fontanillas P, Franco-Cereceda A, Gigante B, Groop L, Gustafsson S, Hager J, Hallmans G, Han BG, Hunt SE, Kang HM, Illig T, Kessler T, Knowles JW, Kolovou G, Kuusisto J, Langenberg C, Langford C, Leander K, Lokki ML, Lundmark A, McCarthy MI, Meisinger C, Melander O, Mihailov E, Maouche S, Morris AD, Müller-Nurasyid M; MuTHER Consortium; Nikus K, Peden JF, Rayner NW, Rasheed A, Rosinger S, Rubin D, Rumpf MP, Schäfer A, Sivananthan M, Song C, Stewart AF, Tan ST, Thorgeirsson G, van der Schoot CE, Wagner PJ; Wellcome Trust Case Control Consortium; Wells GA, Wild PS, Yang TP, Amouyel P, Arveiler D, Basart H, Boehnke M, Boerwinkle E, Brambilla P, Cambien F, Cupples AL, de Faire U, Dehghan A, Diemert P, Epstein SE, Evans A, Ferrario MM, Ferrières J, Gauguier D, Go AS, Goodall AH, Gudnason V, Hazen SL, Holm H, Iribarren C, Jang Y, Kähönen M, Kee F, Kim HS, Klopp N, Koenig W, Kratzer W, Kuulasmaa K, Laakso M, Laaksonen R, Lee JY, Lind L, Ouwehand WH, Parish S, Park JE, Pedersen NL, Peters A, Quertermous T, Rader DJ, Salomaa V, Schadt E, Shah SH, Sinisalo J, Stark K, Stefansson K, Trégouët DA, Virtamo J, Wallentin L, Wareham N, Zimmermann ME, Nieminen MS, Hengstenberg C, Sandhu MS, Pastinen T, Syvänen AC, Hovingh GK, Dedoussis G, Franks PW, Lehtimäki T, Metspalu A, Zalloua PA, Siegbahn A, Schreiber S, Ripatti S, Blankenberg SS, Perola M, Clarke R, Boehm BO, O'Donnell C, Reilly MP, März W, Collins R, Kathiresan S, Hamsten A, Kooner JS, Thorsteinsdottir U, Danesh J, Palmer CN, Roberts R, Watkins H, Schunkert H, Samani NJ. Large-scale association analysis identifies new risk loci for coronary artery disease. *Nat Genet* 2013;**45**:25–33.
131. International Stroke Genetics Consortium (ISGC); Wellcome Trust Case Control Consortium 2 (WTCCC2); Bellenguez C, Bevan S, Gschwendtner A, Spencer CC, Burgess AI, Pirinen M, Jackson CA, Traylor M, Strange A, Su Z, Band G, Syme PD, Malik R, Pera J, Norrving B, Lemmens R, Freeman C, Schanz R, James T, Poole D, Murphy L, Segal H, Cortellini L, Cheng YC, Woo D, Nalls MA, Müller-Myhsok B, Meisinger C, Seedorf U, Ross-Adams H, Boonen S, Wloch-Kopec D, Valant V, Slark J, Furie K, Delavaran H, Langford C, Deloukas P, Edkins S, Hunt S, Gray E, Dronov S, Peltonen L, Gretarsdottir S, Thorleifsson G, Thorsteinsdottir U, Stefansson K, Boncoraglio GB, Parati EA, Attia J, Holliday E, Levi C, Franzosi MG, Goel A, Helgadottir A, Blackwell JM, Bramon E, Brown MA, Casas JP, Corvin A, Duncanson A, Jankowski J, Mathew CG, Palmer CN, Plomin R, Rautanen A, Sawcer SJ, Trembath RC, Viswanathan AC, Wood NW, Worrall BB, Kittner SJ, Mitchell BD, Kissela B, Meschia JF, Thijs V, Lindgren A, Macleod MJ, Slowik A, Walters M, Rosand J, Sharma P, Farrall M, Sudlow CL, Rothwell PM, Dichgans M, Donnelly P, Markus HS. Genome-wide association study identifies a variant in HDAC9 associated with large vessel ischemic stroke. *Nat Genet* 2012;**44**:328–333.
132. Cao Q, Rong S, Repa JJ, St Clair R, Parks JS, Mishra N. Histone deacetylase 9 represses cholesterol efflux and alternatively activated macrophages in atherosclerosis development. *Arterioscler Thromb Vasc Biol* 2014;**34**:1871–1879.
133. Smith-Vikos T, Liu Z, Parsons C, Gorospe M, Ferrucci L, Gill TM, Slack FJ. A serum miRNA profile of human longevity: findings from the Baltimore Longitudinal Study of Aging (BLSA). *Aging (Albany NY)* 2016;**8**:2971–2987.

134. Huan T, Chen G, Liu C, Bhattacharya A, Rong J, Chen BH, Seshadri S, Tanriverdi K, Freedman JE, Larson MG, Murabito JM, Levy D. Age-associated microRNA expression in human peripheral blood is associated with all-cause mortality and age-related traits. *Aging Cell* 2018;**17**:e12687.
135. Marttila S, Chatsirisupachai K, Palmer D, de Magalhaes JP. Ageing-associated changes in the expression of lncRNAs in human tissues reflect a transcriptional modulation in ageing pathways. *Mech Ageing Dev* 2020;**185**:111177.
136. Haque S, Ames RM, Moore K, Pilling LC, Peters LL, Bandinelli S, Ferrucci L, Harries LW. circRNAs expressed in human peripheral blood are associated with human aging phenotypes, cellular senescence and mouse lifespan. *Geroscience* 2020;**42**:183–199.
137. Jusic A, Thomas PB, Wettinger SB, Dogan S, Farrugia R, Gaetano C, Tuna BG, Pinet F, Robinson EL, Tual-Chalot S, Stellos K, Devaux Y; EU-CardioRNA COST Action CA17129. Noncoding RNAs in age-related cardiovascular diseases. *Ageing Res Rev* 2022;**77**:101610.
138. Ito T, Yagi S, Yamakuchi M. MicroRNA-34a regulation of endothelial senescence. *Biochem Biophys Res Commun* 2010;**398**:735–740.
139. Badi I, Burba I, Ruggeri C, Zeni F, Bertolotti M, Scopece A, Pompilio G, Raucci A. MicroRNA-34a induces vascular smooth muscle cells senescence by SIRT1 downregulation and promotes the expression of age-associated pro-inflammatory secretory factors. *J Gerontol A Biol Sci Med Sci* 2015;**70**:1304–1311.
140. Gatsiou A, Georgiopoulos G, Vlachogiannis NI, Pfisterer L, Fischer A, Sachse M, Laina A, Bonini F, Delialis D, Tual-Chalot S, Zormpas E, Achangwa R, Jiang L, Kontogiannis C, Patras R, Hermeking H, Zeiher AM, Stamatelopoulos K, Dimmeler S, Stellos K. Additive contribution of microRNA-34a/b/c to human arterial ageing and atherosclerosis. *Atherosclerosis* 2021;**327**:49–58.
141. Aivalioti E, Georgiopoulos G, Tual-Chalot S, Bampatsias D, Delialis D, Sopova K, Drakos SG, Stellos K, Stamatelopoulos K. Amyloid-beta metabolism in age-related neurocardiovascular diseases. *Eur Heart J* 2024;**46**:250–272.
142. Stakos DA, Stamatelopoulos K, Bampatsias D, Sachse M, Zormpas E, Vlachogiannis NI, Tual-Chalot S, Stellos K. The Alzheimer's disease amyloid-beta hypothesis in cardiovascular aging and disease: JACC focus seminar. *J Am Coll Cardiol* 2020;**75**:952–967.
143. Bampatsias D, Mavroeidis I, Tual-Chalot S, Vlachogiannis NI, Bonini F, Sachse M, Mavraganis G, Mareti A, Kritsioti C, Laina A, Delialis D, Ciliberti G, Sopova K, Gatsiou A, Martelli F, Georgiopoulos G, Stellos K, Stamatelopoulos K. Beta-secretase-1 antisense RNA is associated with vascular ageing and atherosclerotic cardiovascular disease. *Thromb Haemost* 2022;**122**:1932–1942.
144. Sachse M, Tual-Chalot S, Ciliberti G, Amponsah-Offeh M, Stamatelopoulos K, Gatsiou A, Stellos K. RNA-binding proteins in vascular inflammation and atherosclerosis. *Atherosclerosis* 2023;**374**:55–73.
145. Gatsiou A, Tual-Chalot S, Napoli M, Ortega-Gomez A, Regen T, Badolia R, Cesarini V, Garcia-Gonzalez C, Chevre R, Ciliberti G, Silvestre-Roig C, Martini M, Hoffmann J, Hamouche R, Visker JR, Diakos N, Wietelmann A, Silvestris DA, Georgiopoulos G, Moshfegh A, Schneider A, Chen W, Guenther S, Backs J, Kwak S, Selzman CH, Stamatelopoulos K, Rose-John S, Trautwein C, Spyridopoulos I, Braun T, Waisman A, Gallo A, Drakos SG, Dimmeler S, Sperandio M, Soehnlein O, Stellos K. The RNA editor ADAR2 promotes immune cell trafficking by enhancing endothelial responses to interleukin-6 during sterile inflammation. *Immunity* 2023;**56**:979–997.e11.
146. Dorn LE, Tual-Chalot S, Stellos K, Accornero F. RNA epigenetics and cardiovascular diseases. *J Mol Cell Cardiol* 2019;**129**:272–280.
147. Stellos K, Gatsiou A, Stamatelopoulos K, Perisic Matic L, John D, Lunella FF, Jaé N, Rossbach O, Amrhein C, Sigala F, Boon RA, Fürtig B, Manavski Y, You X, Uchida S, Keller T, Boeckel JN, Franco-Cereceda A, Maegdefessel L, Chen W, Schwalbe H, Bindereif A, Eriksson P, Hedin U, Zeiher AM, Dimmeler S. Adenosine-to-inosine RNA editing controls cathepsin S expression in atherosclerosis by enabling HuR-mediated post-transcriptional regulation. *Nat Med* 2016;**22**:1140–1150.
148. Liberale L, Tual-Chalot S, Sedej S, Ministrini S, Georgiopoulos G, Grunewald M, Bäck M, Bochaton-Piallat ML, Boon RA, Ramos GC, de Winther MPJ, Drosatos K, Evans PC, Ferguson JF, Forslund-Startceva SK, Goettsch C, Giacca M, Haendeler J, Kallikourdis M, Ketelhuth DFJ, Koenen RR, Lacolley P, Lutgens E, Maffia P, Miwa S, Monaco C, Montecucco F, Norata GD, Osto E, Richardson GD, Riksen NP, Soehnlein O, Spyridopoulos I, Van Linthout S, Vilahur G, Wentzel JJ, Andrés V, Badimon L, Benetos A, Binder CJ, Brandes RP, Crea F, Furman D, Gorbunova V, Guzik TJ, Hill JA, Lüscher TF, Mittelbrunn M, Nencioni A, Netea MG, Passos JF, Stamatelopoulos KS, Tavernarakis N, Ungvari Z, Wu JC, Kirkland JL, Camici GG, Dimmeler S, Kroemer G, Abdellatif M, Stellos K. Roadmap for alleviating the manifestations of ageing in the cardiovascular system. *Nat Rev Cardiol* 2025;**22**:577–605.
149. Paneni F, Diaz Cañestro C, Libby P, Lüscher TF, Camici GG. The aging cardiovascular system: understanding it at the cellular and clinical levels. *J Am Coll Cardiol* 2017;**69**:1952–1967.
150. Wils J, Djerada Z, Roca F, Duflot T, Iacob M, Remy-Jouet I, Joannides R, Bellien J. Alteration in the availability of epoxyeicosatrienoic acids contributes with NO to the development of endothelial dysfunction in conduit arteries during aging. *Atherosclerosis* 2018;**275**:239–245.
151. Han Y, Kim SY. Endothelial senescence in vascular diseases: current understanding and future opportunities in senotherapeutics. *Exp Mol Med* 2023;**55**:1–12.
152. Ungvari Z, Tarantini S, Kiss T, Wren JD, Giles CB, Griffin CT, Murfee WL, Pacher P, Csiszar A. Endothelial dysfunction and angiogenesis impairment in the ageing vasculature. *Nat Rev Cardiol* 2018;**15**:555–565.
153. Camici GG, Savarese G, Akhmedov A, Lüscher TF. Molecular mechanism of endothelial and vascular aging: implications for cardiovascular disease. *Eur Heart J* 2015;**36**:3392–3403.
154. Karnewar S, Pulipaka S, Katta S, Panuganti D, Neeli PK, Thennati R, Jerald MK, Kotamraju S. Mitochondria-targeted esculetin mitigates atherosclerosis in the setting of aging via the modulation of SIRT1-mediated vascular cell senescence and mitochondrial function in Apoe(-/-) mice. *Atherosclerosis* 2022;**356**:28–40.
155. Karnewar S, Vasamsetti SB, Gopoju R, Kanugula AK, Ganji SK, Prabhakar S, Rangaraj N, Tupperwar N, Kumar JM, Kotamraju S. Mitochondria-targeted esculetin alleviates mitochondrial dysfunction by AMPK-mediated nitric oxide and SIRT3 regulation in endothelial cells: potential implications in atherosclerosis. *Sci Rep* 2016;**6**:24108.
156. Wang JC, Bennett M. Aging and atherosclerosis: mechanisms, functional consequences, and potential therapeutics for cellular senescence. *Circ Res* 2012;**111**:245–259.
157. Galkina E, Ley K. Vascular adhesion molecules in atherosclerosis. *Arterioscler Thromb Vasc Biol* 2007;**27**:2292–2301.
158. Li X, Li C, Zhang W, Wang Y, Qian P, Huang H. Inflammation and aging: signaling pathways and intervention therapies. *Signal Transduct Target Ther* 2023;**8**:239.
159. Monti LD, Barlassina C, Citterio L, Galluccio E, Berzuini C, Setola E, Valsecchi G, Lucotti P, Pozza G, Bernardinelli L, Casari G, Piatti P. Endothelial nitric oxide synthase polymorphisms are associated with type 2 diabetes and the insulin resistance syndrome. *Diabetes* 2003;**52**:1270–1275.
160. Bondareva O, Rodríguez-Aguilera JR, Oliveira F, Liao L, Rose A, Gupta A, Singh K, Geier F, Schuster J, Boeckel JN, Buescher JM, Kohli S, Klöting N, Isermann B, Blüher M, Sheikh BN. Single-cell profiling of vascular endothelial cells reveals progressive organ-specific vulnerabilities during obesity. *Nat Metab* 2022;**4**:1591–1610.
161. Suzuki K, Hatzikotoulas K, Southam L, Taylor HJ, Yin X, Lorenz KM, Mandla R, Huerta-Chagoya A, Melloni GEM, Kanoni S, Rayner NW, Bocher O, Arruda AL, Sonehara K, Namba S, Lee SSK, Preuss MH, Petty LE, Schroeder P, Vanderwerff B, Kals M, Bragg F, Lin K, Guo X, Zhang W, Yao J, Kim YJ, Graff M, Takeuchi F, Nano J, Lamri A, Nakatochi M, Moon S, Scott RA, Cook JP, Lee JJ, Pan I, Taliun D, Parra EJ, Chai JF, Bielak LF, Tabara Y, Hai Y, Thorleifsson G, Grarup N, Sofer T, Wuttke M, Sarnowski C, Gieger C, Nousome D, Trompet S, Kwak SH, Long J, Sun M, Tong L, Chen WM, Nongmaithem SS, Noordam R, Lim VJY, Tam CHT, Joo YY, Chen CH, Raffield LM, Prins BP, Nicolas A, Yanek LR, Chen G, Brody JA, Kabagambe E, An P, Xiang AH, Choi HS, Cade BE, Tan J, Broadaway KA, Williamson A, Kamali Z, Cui J, Thangam M, Adair LS, Adeyemo A, Aguilar-Salinas CA, Ahluwalia TS, Anand SS, Bertoni A, Bork-Jensen J, Brandslund I, Buchanan TA, Burant CF, Butterworth AS, Canouil M, Chan JCN, Chang LC, Chee ML, Chen J, Chen SH, Chen YT, Chen Z, Chuang LM, Cushman M, Danesh J, Das SK, de Silva HJ, Dedoussis G, Dimitrov L, Doumatey AP, Du S, Duan Q, Eckardt KU, Emery LS, Evans DS, Evans MK, Fischer K, Floyd JS, Ford I, Franco OH, Frayling TM, Freedman BI, Genter P, Gerstein HC, Giedraitis V, González-Villalpando C, González-Villalpando ME, Gordon-Larsen P, Gross M, Guare LA, Hackinger S, Hakaste L, Han S, Hattersley AT, Herder C, Horikoshi M, Howard AG, Hsueh W, Huang M, Huang W, Hung YJ, Hwang MY, Hwu CM, Ichihara S, Ikram MA, Ingelsson M, Islam MT, Isono M, Jang HM, Jasmine F, Jiang G, Jonas JB, Jørgensen T, Kamanu FK, Kandeel FR, Kasturiratne A, Katsuya T, Kaur V, Kawaguchi T, Keaton JM, Kho AN, Khor CC, Kibriya MG, Kim DH, Kronenberg F, Kuusisto J, Läll K, Lange LA, Lee KM, Lee MS, Lee NR, Leong A, Li L, Li Y, Li-Gao R, Ligthart S, Lindgren CM, Linneberg A, Liu CT, Liu J, Locke AE, Louie T, Luan J, Luk AO, Luo X, Lv J, Lynch JA, Lyssenko V, Maeda S, Mamakou V, Mansuri SR, Matsuda K, Meitinger T, Melander O, Metspalu A, Mo H, Morris AD, Moura FA, Nadler JL, Nalls MA, Nayak U, Ntalla I, Okada Y, Orozco L, Patel SR, Patil S, Pei P, Pereira MA, Peters A, Pirie FJ, Polikowsky HG, Porneala B, Prasad G, Rasmussen-Torvik LJ, Reiner AP, Roden M, Rohde R, Roll K, Sabanayagam C, Sandow K, Sankareswaran A, Sattar N, Schönherr S, Shahriar M, Shen B, Shi J, Shin DM, Shojima N, Smith JA, So WY, Stančáková A, Steinthorsdottir V, Stilp AM, Strauch K, Taylor KD, Thorand B, Thorsteinsdottir U, Tomlinson B, Tran TC, Tsai FJ, Tuomilehto J, Tusie-Luna T, Udler MS, Valladares-Salgado A, van Dam RM, van Klinken JB, Varma R, Wacher-Rodarte N, Wheeler E, Wickremasinghe AR, van Dijk KW, Witte DR, Yajnik CS, Yamamoto K, Yamamoto K, Yoon K, Yu C, Yuan JM, Yusuf S, Zawistowski M, Zhang L, Zheng W; VA Million Veteran Program; Raffel LJ, Igase M, Ipp E, Redline S, Cho YS, Lind L, Province MA, Fornage M, Hanis CL, Ingelsson E, Zonderman AB, Psaty BM, Wang YX, Rotimi CN, Becker DM, Matsuda F, Liu Y, Yokota M, Kardia SLR, Peyser PA, Pankow JS, Engert JC, Bonnefond A, Froguel P, Wilson JG, Sheu WHH, Wu JY, Hayes MG, Ma RCW, Wong TY, Mook-Kanamori DO, Tuomi T, Chandak GR, Collins FS, Bharadwaj D, Paré G, Sale MM, Ahsan H, Motala AA, Shu XO, Park KS, Jukema JW, Cruz M, Chen YI, Rich SS, McKean-Cowdin R, Grallert H, Cheng CY, Ghanbari M, Tai ES, Dupuis J, Kato N, Laakso M, Köttgen A, Koh WP, Bowden DW, Palmer CNA, Kooner JS, Kooperberg C, Liu S, North KE, Saleheen D, Hansen T, Pedersen O, Wareham NJ, Lee J, Kim BJ, Millwood IY, Walters RG, Stefansson K, Ahlqvist E, Goodarzi MO, Mohlke KL, Langenberg C, Haiman CA, Loos RJF, Florez JC, Rader DJ, Ritchie MD, Zöllner S, Mägi R, Marston NA, Ruff CT, van Heel DA, Finer S, Denny JC, Yamauchi T, Kadowaki T, Chambers JC, Ng MCY, Sim X, Below JE, Tsao PS, Chang KM, McCarthy MI, Meigs JB, Mahajan A, Spracklen CN, Mercader JM, Boehnke M, Rotter JI, Vujkovic M, Voight BF, Morris AP, Zeggini E. Genetic drivers of heterogeneity in type 2 diabetes pathophysiology. *Nature* 2024;**627**:347–357.
162. Donato AJ, Black AD, Jablonski KL, Gano LB, Seals DR. Aging is associated with greater nuclear NF kappa B, reduced I kappa B alpha, and increased expression of proinflammatory cytokines in vascular endothelial cells of healthy humans. *Aging Cell* 2008;**7**:805–812.
163. Donato AJ, Eskurza I, Silver AE, Levy AS, Pierce GL, Gates PE, Seals DR. Direct evidence of endothelial oxidative stress with aging in humans: relation to impaired endothelium-

dependent dilation and upregulation of nuclear factor-kappaB. *Circ Res* 2007;**100**:1659–1666.
164. Lacolley P, Regnault V, Segers P, Laurent S. Vascular smooth muscle cells and arterial stiffening: relevance in development, aging, and disease. *Physiol Rev* 2017;**97**:1555–1617.
165. Ma Z, Mao C, Jia Y, Fu Y, Kong W. Extracellular matrix dynamics in vascular remodeling. *Am J Physiol Cell Physiol* 2020;**319**:C481–c499.
166. Propson NE, Roy ER, Litvinchuk A, Köhl J, Zheng H. Endothelial C3a receptor mediates vascular inflammation and blood-brain barrier permeability during aging. *J Clin Invest* 2021;**131**:e140966.
167. Katsimpardi L, Litterman NK, Schein PA, Miller CM, Loffredo FS, Wojtkiewicz GR, Chen JW, Lee RT, Wagers AJ, Rubin LL. Vascular and neurogenic rejuvenation of the aging mouse brain by young systemic factors. *Science* 2014;**344**:630–634.
168. Jacob MP. Extracellular matrix remodeling and matrix metalloproteinases in the vascular wall during aging and in pathological conditions. *Biomed Pharmacother* 2003;**57**:195–202.
169. Lanzer P, Hannan FM, Lanzer JD, Janzen J, Raggi P, Furniss D, Schuchardt M, Thakker R, Fok PW, Saez-Rodriguez J, Millan A, Sato Y, Ferraresi R, Virmani R, St Hilaire C. Medial arterial calcification: JACC State-of-the-Art Review. *J Am Coll Cardiol* 2021;**78**:1145–1165.
170. Said MA, Eppinga RN, Lipsic E, Verweij N, van der Harst P. Relationship of Arterial Stiffness Index and pulse pressure with cardiovascular disease and mortality. *J Am Heart Assoc* 2018;**7**:e007621.
171. Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. *N Engl J Med* 2005;**352**:1685–1695.
172. Libby P. The changing landscape of atherosclerosis. *Nature* 2021;**592**:524–533.
173. Gimbrone MA J, García-Cardeña G. Endothelial cell dysfunction and the pathobiology of atherosclerosis. *Circ Res* 2016;**118**:620–636.
174. Souilhol C, Serbanovic-Canic J, Fragiadaki M, Chico TJ, Ridger V, Roddie H, Evans PC. Endothelial responses to shear stress in atherosclerosis: a novel role for developmental genes. *Nat Rev Cardiol* 2020;**17**:52–63.
175. Smeets MWJ, Mourik MJ, Niessen HWM, Hordijk PL. Stasis promotes erythrocyte adhesion to von willebrand factor. *Arterioscler Thromb Vasc Biol* 2017;**37**:1618–1627.
176. Monos E, Lóránt M, Dörnyei G, Bérczi V, Nádasy G. Long-term adaptation mechanisms in extremity veins supporting orthostatic tolerance. *News Physiol Sci* 2003;**18**:210–214.
177. Cui Y, Gollasch M, Kassmann M. Arterial myogenic response and aging. *Ageing Res Rev* 2023;**84**:101813.
178. Jakovljevic DG. Physical activity and cardiovascular aging: physiological and molecular insights. *Exp Gerontol* 2018;**109**:67–74.
179. Shannon OM, Mendes I, Köchl C, Mazidi M, Ashor AW, Rubele S, Minihane AM, Mathers JC, Siervo M. Mediterranean diet increases endothelial function in adults: a systematic review and meta-analysis of randomized controlled trials. *J Nutr* 2020;**150**:1151–1159.
180. Dupuis J, Tardif JC, Cernacek P, Théroux P. Cholesterol reduction rapidly improves endothelial function after acute coronary syndromes. The RECIFE (reduction of cholesterol in ischemia and function of the endothelium) trial. *Circulation* 1999;**99**:3227–3233.
181. Silva IVG, de Figueiredo RC, Rios DRA. Effect of different classes of antihypertensive drugs on endothelial function and inflammation. *Int J Mol Sci* 2019;**20**:3458.
182. John CM, Khaddaj Mallat R, Mishra RC, George G, Singh V, Turnbull JD, Umeshappa CS, Kendrick DJ, Kim T, Fauzi FM, Visser F, Fedak PWM, Wulff H, Braun AP. SKA-31, an activator of Ca(2+)-activated K(+) channels, improves cardiovascular function in aging. *Pharmacol Res* 2020;**151**:104539.
183. Zhu Y, Tchkonia T, Pirtskhalava T, Gower AC, Ding H, Giorgadze N, Palmer AK, Ikeno Y, Hubbard GB, Lenburg M, O'Hara SP, LaRusso NF, Miller JD, Roos CM, Verzosa GC, LeBrasseur NK, Wren JD, Farr JN, Khosla S, Stout MB, McGowan SJ, Fuhrmann-Stroissnigg H, Gurkar AU, Zhao J, Colangelo D, Dorronsoro A, Ling YY, Barghouthy AS, Navarro DC, Sano T, Robbins PD, Niedernhofer LJ, Kirkland JL. The Achilles' heel of senescent cells: from transcriptome to senolytic drugs. *Aging Cell* 2015;**14**:644–658.
184. Zhu Y, Tchkonia T, Fuhrmann-Stroissnigg H, Dai HM, Ling YY, Stout MB, Pirtskhalava T, Giorgadze N, Johnson KO, Giles CB, Wren JD, Niedernhofer LJ, Robbins PD, Kirkland JL. Identification of a novel senolytic agent, navitoclax, targeting the Bcl-2 family of anti-apoptotic factors. *Aging Cell* 2016;**15**:428–435.
185. Assar M E, Angulo J, Rodríguez-Mañas L. Oxidative stress and vascular inflammation in aging. *Free Radic Biol Med* 2013;**65**:380–401.
186. Chen YR, Zweier JL. Cardiac mitochondria and reactive oxygen species generation. *Circ Res* 2014;**114**:524–537.
187. Muntean DM, Sturza A, Dănilă MD, Borza C, Duicu OM, Mornoș C. The role of mitochondrial reactive oxygen Species in cardiovascular injury and protective strategies. *Oxid Med Cell Longev* 2016;**2016**:8254942.
188. Gómez J, Mota-Martorell N, Jové M, Pamplona R, Barja G. Mitochondrial ROS production, oxidative stress and aging within and between species: evidences and recent advances on this aging effector. *Exp Gerontol* 2023;**174**:112134.
189. Rodríguez-Mañas L, El-Assar M, Vallejo S, López-Dóriga P, Solís J, Petidier R, Montes M, Nevado J, Castro M, Gómez-Guerrero C, Peiró C, Sánchez-Ferrer CF. Endothelial dysfunction in aged humans is related with oxidative stress and vascular inflammation. *Aging Cell* 2009;**8**:226–238.
190. Harman D. Aging: a theory based on free radical and radiation chemistry. *J Gerontol* 1956;**11**:298–300.
191. Deepa SS, Van Remmen H, Brooks SV, Faulkner JA, Larkin L, McArdle A, Jackson MJ, Vasilaki A, Richardson A. Accelerated sarcopenia in Cu/Zn superoxide dismutase knockout mice. *Free Radic Biol Med* 2019;**132**:19–23.
192. Muller FL, Song W, Liu Y, Chaudhuri A, Pieke-Dahl S, Strong R, Huang TT, Epstein CJ, Roberts LJ 2nd, Csete M, Faulkner JA, Van Remmen H. Absence of CuZn superoxide dismutase leads to elevated oxidative stress and acceleration of age-dependent skeletal muscle atrophy. *Free Radic Biol Med* 2006;**40**:1993–2004.
193. Jang YC, Lustgarten MS, Liu Y, Muller FL, Bhattacharya A, Liang H, Salmon AB, Brooks SV, Larkin L, Hayworth CR, Richardson A, Van Remmen H. Increased superoxide in vivo accelerates age-associated muscle atrophy through mitochondrial dysfunction and neuromuscular junction degeneration. *FASEB J* 2010;**24**:1376–1390.
194. Li Y, Huang TT, Carlson EJ, Melov S, Ursell PC, Olson JL, Noble LJ, Yoshimura MP, Berger C, Chan PH, Wallace DC, Epstein CJ. Dilated cardiomyopathy and neonatal lethality in mutant mice lacking manganese superoxide dismutase. *Nat Genet* 1995;**11**:376–381.
195. Wenzel P, Schuhmacher S, Kienhöfer J, Müller J, Hortmann M, Oelze M, Schulz E, Treiber N, Kawamoto T, Scharffetter-Kochanek K, Münzel T, Bürkle A, Bachschmid MM, Daiber A. Manganese superoxide dismutase and aldehyde dehydrogenase deficiency increase mitochondrial oxidative stress and aggravate age-dependent vascular dysfunction. *Cardiovasc Res* 2008;**80**:280–289.
196. Oelze M, Kröller-Schön S, Steven S, Lubos E, Doppler C, Hausding M, Tobias S, Brochhausen C, Li H, Torzewski M, Wenzel P, Bachschmid M, Lackner KJ, Schulz E, Münzel T, Daiber A. Glutathione peroxidase-1 deficiency potentiates dysregulatory modifications of endothelial nitric oxide synthase and vascular dysfunction in aging. *Hypertension* 2014;**63**:390–396.
197. Espinola-Klein C, Rupprecht HJ, Bickel C, Schnabel R, Genth-Zotz S, Torzewski M, Lackner K, Munzel T, Blankenberg S; AtheroGene Investigators. Glutathione peroxidase-1 activity, atherosclerotic burden, and cardiovascular prognosis. *Am J Cardiol* 2007;**99**:808–812.
198. Blankenberg S, Rupprecht HJ, Bickel C, Torzewski M, Hafner G, Tiret L, Smieja M, Cambien F, Meyer J, Lackner KJ; AtheroGene Investigators. Glutathione peroxidase 1 activity and cardiovascular events in patients with coronary artery disease. *N Engl J Med* 2003;**349**:1605–1613.
199. Barja G, Herrero A. Oxidative damage to mitochondrial DNA is inversely related to maximum life span in the heart and brain of mammals. *FASEB J* 2000;**14**:312–318.
200. Matrone G, Thandavarayan RA, Walther BK, Meng S, Mojiri A, Cooke JP. Dysfunction of iPSC-derived endothelial cells in human Hutchinson-Gilford progeria syndrome. *Cell Cycle* 2019;**18**:2495–2508.
201. Bidault G, Garcia M, Capeau J, Morichon R, Vigouroux C, Béréziat V. Progerin expression induces inflammation, oxidative stress and senescence in human coronary endothelial cells. *Cells* 2020;**9**:1201.
202. Atchison L, Abutaleb NO, Snyder-Mounts E, Gete Y, Ladha A, Ribar T, Cao K, Truskey GA. iPSC-derived endothelial cells affect vascular function in a tissue-engineered blood vessel model of Hutchinson-Gilford Progeria syndrome. *Stem Cell Reports* 2020;**14**:325–337.
203. Brandt M, Dörschmann H, Khraisat S, Knopp T, Ringen J, Kalinovic S, Garlapati V, Siemer S, Molitor M, Göbel S, Stauber R, Karbach SH, Münzel T, Daiber A, Wenzel P. Telomere shortening in hypertensive heart disease depends on oxidative DNA damage and predicts impaired recovery of cardiac function in heart failure. *Hypertension* 2022;**79**:2173–2184.
204. Schmidt-Trucksäss A, Lichtenstein AH, von Känel R. Lifestyle factors as determinants of atherosclerotic cardiovascular health. *Atherosclerosis* 2024;**395**:117577.
205. Mone P, Varzideh F, Jankauskas SS, Pansini A, Lombardi A, Frullone S, Santulli G. SGLT2 inhibition via empagliflozin improves endothelial function and reduces mitochondrial oxidative stress: insights from frail hypertensive and diabetic patients. *Hypertension* 2022;**79**:1633–1643.
206. Park SH, Belcastro E, Hasan H, Matsushita K, Marchandot B, Abbas M, Toti F, Auger C, Jesel L, Ohlmann P, Morel O, Schini-Kerth VB. Angiotensin II-induced upregulation of SGLT1 and 2 contributes to human microparticle-stimulated endothelial senescence and dysfunction: protective effect of gliflozins. *Cardiovasc Diabetol* 2021;**20**:65.
207. Mohammed I, Hollenberg MD, Ding H, Triggle CR. A critical review of the evidence that metformin is a putative anti-aging drug that enhances healthspan and extends lifespan. *Front Endocrinol (Lausanne)* 2021;**12**:718942.
208. Santulli G, Kansakar U, Varzideh F, Mone P, Jankauskas SS, Lombardi A. Functional role of taurine in aging and cardiovascular health: an updated overview. *Nutrients* 2023;**15**:4236.
209. Franceschi C, Bonafè M, Valensin S, Olivieri F, De Luca M, Ottaviani E, De Benedictis G. Inflamm-aging. An evolutionary perspective on immunosenescence. *Ann N Y Acad Sci* 2000;**908**:244–254.
210. Gardner ID. The effect of aging on susceptibility to infection. *Rev Infect Dis* 1980;**2**:801–810.
211. Ershler WB. Interleukin-6: a cytokine for gerontologists. *J Am Geriatr Soc* 1993;**41**:176–181.
212. Goronzy JJ, Weyand CM. Immune aging and autoimmunity. *Cell Mol Life Sci* 2012;**69**:1615–1623.
213. Liu Z, Liang Q, Ren Y, Guo C, Ge X, Wang L, Cheng Q, Luo P, Zhang Y, Han X. Immunosenescence: molecular mechanisms and diseases. *Signal Transduct Target Ther* 2023;**8**:200.
214. Shaw AC, Goldstein DR, Montgomery RR. Age-dependent dysregulation of innate immunity. *Nat Rev Immunol* 2013;**13**:875–887.
215. Dillon K, Goodman Zachary T, Kaur Sonya S, Levin B, McIntosh R. Neutrophil-to-lymphocyte ratio amplifies the effects of aging on decrements in grip strength and its functional neural underpinnings. *J Gerontol A Biol Sci Med Sci* 2023;**78**:882–889.
216. He Y, Liu X, Wang M, Ke H, Ge C. Neutrophil-to-lymphocyte ratio as a predictor of cardiovascular mortality in cancer survivors. *Sci Rep* 2024;**14**:20980.

217. Galardo G, Crisanti L, Gentile A, Cornacchia M, Iatomasi F, Egiddi I, Puscio E, Menichelli D, Pugliese F, Pastori D; and the Research on Medical patients Admitted to the Emergency Department (ROMA-ED) investigators. Neutrophil to lymphocyte ratio (NLR) and short-term mortality risk in elderly acute medical patients admitted to a University Hospital Emergency Department. *Intern Emerg Med* 2025;**20**:553–562.
218. Cupido AJ, Kraaijenhof JM, Burgess S, Asselbergs FW, Hovingh GK, Gill D. Genetically predicted neutrophil-to-lymphocyte ratio and coronary artery disease: evidence from Mendelian randomization. *Circ Genom Precis Med* 2022;**15**:e003553.
219. Ridker PM, Moorthy MV, Cook NR, Rifai N, Lee I-M, Buring JE. Inflammation, cholesterol, lipoprotein(a), and 30-year cardiovascular outcomes in women. *N Engl J Med* 2024;**391**: 2087–2097.
220. Drosos GC, Vedder D, Houben E, Boekel L, Atzeni F, Badreh S, Boumpas DT, Brodin N, Bruce IN, González-Gay MÁ, Jacobsen S, Kerekes G, Marchiori F, Mukhtyar C, Ramos-Casals M, Sattar N, Schreiber K, Sciascia S, Svenungsson E, Szekanecz Z, Tausche AK, Tyndall A, van Halm V, Voskuyl A, Macfarlane GJ, Ward MM, Nurmohamed MT, Tektonidou MG. EULAR recommendations for cardiovascular risk management in rheumatic and musculoskeletal diseases, including systemic lupus erythematosus and antiphospholipid syndrome. *Ann Rheum Dis* 2022;**81**:768–779.
221. Tardif JC, Kouz S, Waters DD, Bertrand OF, Diaz R, Maggioni AP, Pinto FJ, Ibrahim R, Gamra H, Kiwan GS, Berry C, López-Sendón J, Ostadal P, Koenig W, Angoulvant D, Grégoire JC, Lavoie MA, Dubé MP, Rhainds D, Provencher M, Blondeau L, Orfanos A, L'Allier PL, Guertin MC, Roubille F. Efficacy and safety of low-dose colchicine after myocardial infarction. *N Engl J Med* 2019;**381**:2497–2505.
222. Nidorf SM, Fiolet ATL, Mosterd A, Eikelboom JW, Schut A, Opstal TSJ, The SHK, Xu XF, Ireland MA, Lenderink T, Latchem D, Hoogslag P, Jerzewski A, Nierop P, Whelan A, Hendriks R, Swart H, Schaap J, Kuijper AFM, van Hessen MWJ, Saklani P, Tan I, Thompson AG, Morton A, Judkins C, Bax WA, Dirksen M, Alings M, Hankey GJ, Budgeon CA, Tijssen JGP, Cornel JH, Thompson PL; LoDoCo2 Trial Investigators. Colchicine in patients with chronic coronary disease. *N Engl J Med* 2020;**383**:1838–1847.
223. Fiolet ATL, Poorthuis MHF, Opstal TSJ, Amarenco P, Boczar KE, Buysschaert I, Budgeon C, Chan NC, Cornel JH, Jolly SS, Layland J, Lemmens R, Mewton N, Nidorf SM, Pascual-Figal DA, Price C, Shah B, Tardif JC, Thompson PL, Tijssen JGP, Tsivgoulis G, Walsh C, Wang Y, Weimar C, Eikelboom JW, Mosterd A, Kelly PJ; Colchicine Cardiovascular Trialists Collaboration. Colchicine for secondary prevention of ischaemic stroke and atherosclerotic events: a meta-analysis of randomised trials. *EClinicalMedicine* 2024;**76**:102835.
224. Bawamia B, Spray L, Wangsaputra VK, Bennaceur K, Vahabi S, Stellos K, Kharatikoopaei E, Ogundimu E, Gale CP, Keavney B, Maier R, Hancock H, Richardson G, Austin D, Spyridopoulos I. Activation of telomerase by TA-65 enhances immunity and reduces inflammation post myocardial infarction. *Geroscience* 2023;**45**:2689–2705.
225. Kiss T, Tarantini S, Csipo T, Balasubramanian P, Nyúl-Tóth Á, Yabluchanskiy A, Wren JD, Garman L, Huffman DM, Csiszar A, Ungvari Z. Circulating anti-geronic factors from heterochronic parabionts promote vascular rejuvenation in aged mice: transcriptional footprint of mitochondrial protection, attenuation of oxidative stress, and rescue of endothelial function by young blood. *Geroscience* 2020;**42**:727–748.
226. Nyúl-Tóth Á, Tarantini S, DelFavero J, Yan F, Balasubramanian P, Yabluchanskiy A, Ahire C, Kiss T, Csipo T, Lipecz A, Farkas AE, Wilhelm I, Krizbai IA, Tang Q, Csiszar A, Ungvari Z. Demonstration of age-related blood-brain barrier disruption and cerebromicrovascular rarefaction in mice by longitudinal intravital two-photon microscopy and optical coherence tomography. *Am J Physiol Heart Circ Physiol* 2021;**320**:H1370–h1392.
227. Cheng S, Fernandes VR, Bluemke DA, McClelland RL, Kronmal RA, Lima JA. Age-related left ventricular remodeling and associated risk for cardiovascular outcomes: the Multi-Ethnic Study of Atherosclerosis. *Circ Cardiovasc Imaging* 2009;**2**:191–198.
228. Toth P, Tarantini S, Csiszar A, Ungvari Z. Functional vascular contributions to cognitive impairment and dementia: mechanisms and consequences of cerebral autoregulatory dysfunction, endothelial impairment, and neurovascular uncoupling in aging. *Am J Physiol Heart Circ Physiol* 2017;**312**:H1–h20.
229. Lembo M, Pacella D, Manzi MV, Morisco C, La Mura L, Mancusi C, Bardi L, Trimarco V, Trimarco B, Izzo R, Esposito G. Hypertension-mediated organ damage involving multiple sites is an independent risk factor for cardiovascular events. *Eur Heart J Open* 2023;**3**: oead102.
230. Robbins JL, Jones WS, Duscha BD, Allen JD, Kraus WE, Regensteiner JG, Hiatt WR, Annex BH. Relationship between leg muscle capillary density and peak hyperemic blood flow with endurance capacity in peripheral artery disease. *J Appl Physiol (1985)* 2011;**111**:81–86.
231. Mullins RF, Schoo DP, Sohn EH, Flamme-Wiese MJ, Workamelahu G, Johnston RM, Wang K, Tucker BA, Stone EM. The membrane attack complex in aging human choriocapillaris: relationship to macular degeneration and choroidal thinning. *Am J Pathol* 2014;**184**: 3142–3153.
232. Guymer RH, Campbell TG. Age-related macular degeneration. *Lancet* 2023;**401**: 1459–1472.
233. LeBlanc AJ, Hoying JB. Adaptation of the coronary microcirculation in aging. *Microcirculation* 2016;**23**:157–167.
234. Osnabrugge RL, Mylotte D, Head SJ, Van Mieghem NM, Nkomo VT, LeReun CM, Bogers AJ, Piazza N, Kappetein AP. Aortic stenosis in the elderly: disease prevalence and number of candidates for transcatheter aortic valve replacement: a meta-analysis and modeling study. *J Am Coll Cardiol* 2013;**62**:1002–1012.
235. Blaser MC, Kraler S, Lüscher TF, Aikawa E. Multi-omics approaches to define calcific aortic valve disease pathogenesis. *Circ Res* 2021;**128**:1371–1397.
236. Bäck M, Michel JB. From organic and inorganic phosphates to valvular and vascular calcifications. *Cardiovasc Res* 2021;**117**:2016–2029.
237. Conrad N, Judge A, Tran J, Mohseni H, Hedgecott D, Crespillo AP, Allison M, Hemingway H, Cleland JG, McMurray JJV, Rahimi K. Temporal trends and patterns in heart failure incidence: a population-based study of 4 million individuals. *Lancet* 2018;**391**:572–580.
238. Strait JB, Lakatta EG. Aging-associated cardiovascular changes and their relationship to heart failure. *Heart Fail Clin* 2012;**8**:143–164.
239. Janczewski AM, Spurgeon HA, Lakatta EG. Action potential prolongation in cardiac myocytes of old rats is an adaptation to sustain youthful intracellular Ca2 + regulation. *J Mol Cell Cardiol* 2002;**34**:641–648.
240. Lakatta EG. Cardiovascular regulatory mechanisms in advanced age. *Physiol Rev* 1993;**73**: 413–467.
241. Fang Z, Raza U, Song J, Lu J, Yao S, Liu X, Zhang W, Li S. Systemic aging fuels heart failure: molecular mechanisms and therapeutic avenues. *ESC Heart Fail* 2025;**12**:1059–1080.
242. Camici PG, Tschöpe C, Di Carli MF, Rimoldi O, Van Linthout S. Coronary microvascular dysfunction in hypertrophy and heart failure. *Cardiovasc Res* 2020;**116**:806–816.
243. Dong M, Yang Z, Fang H, Xiang J, Xu C, Zhou Y, Wu Q, Liu J. Aging attenuates cardiac contractility and affects therapeutic consequences for myocardial infarction. *Aging Dis* 2020;**11**: 365–376.
244. Mogensen UM, Ersbøll M, Andersen M, Andersson C, Hassager C, Torp-Pedersen C, Gustafsson F, Køber L. Clinical characteristics and major comorbidities in heart failure patients more than 85 years of age compared with younger age groups. *Eur J Heart Fail* 2011; **13**:1216–1223.
245. Playford D, Strange G, Celermajer DS, Evans G, Scalia GM, Stewart S, Prior D; NEDA Investigators. Diastolic dysfunction and mortality in 436 360 men and women: the National Echo Database Australia (NEDA). *Eur Heart J Cardiovasc Imaging* 2021;**22**: 505–515.
246. Fouda AY, Fagan SC, Ergul A. Brain vasculature and cognition. *Arterioscler Thromb Vasc Biol* 2019;**39**:593–602.
247. Nagata K, Yamazaki T, Takano D, Maeda T, Fujimaki Y, Nakase T, Sato Y. Cerebral circulation in aging. *Ageing Res Rev* 2016;**30**:49–60.
248. Rabin JS, Schultz AP, Hedden T, Viswanathan A, Marshall GA, Kilpatrick E, Klein H, Buckley RF, Yang HS, Properzi M, Rao V, Kirn DR, Papp KV, Rentz DM, Johnson KA, Sperling RA, Chhatwal JP. Interactive associations of vascular risk and β-amyloid burden with cognitive decline in clinically normal elderly individuals: findings from the harvard aging brain study. *JAMA Neurol* 2018;**75**:1124–1131.
249. Wiseman S, Marlborough F, Doubal F, Webb DJ, Wardlaw J. Blood markers of coagulation, fibrinolysis, endothelial dysfunction and inflammation in lacunar stroke versus non-lacunar stroke and non-stroke: systematic review and meta-analysis. *Cerebrovasc Dis* 2014;**37**: 64–75.
250. Bennett HC, Zhang Q, Wu YT, Manjila SB, Chon U, Shin D, Vanselow DJ, Pi HJ, Drew PJ, Kim Y. Aging drives cerebrovascular network remodeling and functional changes in the mouse brain. *Nat Commun* 2024;**15**:6398.
251. Yang T, Sun Y, Lu Z, Leak RK, Zhang F. The impact of cerebrovascular aging on vascular cognitive impairment and dementia. *Ageing Res Rev* 2017;**34**:15–29.
252. van Elderen SG, de Roos A, de Craen AJ, Westendorp RG, Blauw GJ, Jukema JW, Bollen EL, Middelkoop HA, van Buchem MA, van der Grond J. Progression of brain atrophy and cognitive decline in diabetes mellitus: a 3-year follow-up. *Neurology* 2010;**75**:997–1002.
253. Juul Rasmussen I, Rasmussen KL, Nordestgaard BG, Tybjærg-Hansen A, Frikke-Schmidt R. Impact of cardiovascular risk factors and genetics on 10-year absolute risk of dementia: risk charts for targeted prevention. *Eur Heart J* 2020;**41**:4024–4033.
254. Malik R, Georgakis MK, Neitzel J, Rannikmäe K, Ewers M, Seshadri S, Sudlow CLM, Dichgans M. Midlife vascular risk factors and risk of incident dementia: longitudinal cohort and Mendelian randomization analyses in the UK Biobank. *Alzheimers Dement* 2021;**17**: 1422–1431.
255. Singh-Manoux A, Dugravot A, Shipley M, Brunner EJ, Elbaz A, Sabia S, Kivimaki M. Obesity trajectories and risk of dementia: 28 years of follow-up in the Whitehall II study. *Alzheimers Dement* 2018;**14**:178–186.
256. O'Donnell M, Teo K, Gao P, Anderson C, Sleight P, Dans A, Marzona I, Bosch J, Probstfield J, Yusuf S. Cognitive impairment and risk of cardiovascular events and mortality. *Eur Heart J* 2012;**33**:1777–1786.
257. Schievink SHJ, van Boxtel MPJ, Deckers K, van Oostenbrugge RJ, Verhey FRJ, Köhler S. Cognitive changes in prevalent and incident cardiovascular disease: a 12-year follow-up in the Maastricht Aging Study (MAAS). *Eur Heart J* 2022;**43**:e2–e9.
258. Damluji AA, Chung SE, Xue QL, Hasan RK, Moscucci M, Forman DE, Bandeen-Roche K, Batchelor W, Walston JD, Resar JR, Gerstenblith G. Frailty and cardiovascular outcomes in the National Health and Aging Trends Study. *Eur Heart J* 2021;**42**:3856–3865.
259. Argentieri MA, Amin N, Nevado-Holgado AJ, Sproviero W, Collister JA, Keestra SM, Kuilman MM, Ginos BNR, Ghanbari M, Doherty A, Hunter DJ, Alvergne A, van Duijn CM. Integrating the environmental and genetic architectures of aging and mortality. *Nat Med* 2025;**31**:1016–1025.
260. Franklin SS, Jacobs MJ, Wong ND, L'Italien GJ, Lapuerta P. Predominance of isolated systolic hypertension among middle-aged and elderly US hypertensives: analysis based on National Health and Nutrition Examination Survey (NHANES) III. *Hypertension* 2001;**37**:869–874.
261. Tang KS, Jones JE, Fan W, Wong ND. Prevalence and mortality trends of hypertension subtypes among US adults: an analysis of the National Health and Nutrition Examination Survey (NHANES). *Am J Hypertens* 2025;**38**:303–312.

262. McEvoy JW, Daya N, Rahman F, Hoogeveen RC, Blumenthal RS, Shah AM, Ballantyne CM, Coresh J, Selvin E. Association of isolated diastolic hypertension as defined by the 2017 ACC/AHA blood pressure guideline with incident cardiovascular outcomes. *JAMA* 2020; **323**:329–338.
263. Laurent S, Boutouyrie P. Arterial stiffness and hypertension in the elderly. *Front Cardiovasc Med* 2020;**7**:544302.
264. Satish S, Zhang DD, Goodwin JS. Clinical significance of falling blood pressure among older adults. *J Clin Epidemiol* 2001;**54**:961–967.
265. Rådholm K, Festin K, Falk M, Midlöv P, Mölstad S, Östgren CJ. Blood pressure and all-cause mortality: a prospective study of nursing home residents. *Age Ageing* 2016;**45**:826–832.
266. Streit S, Poortvliet RKE, Gussekloo J. Lower blood pressure during antihypertensive treatment is associated with higher all-cause mortality and accelerated cognitive decline in the oldest-old. Data from the Leiden 85-plus study. *Age Ageing* 2018;**47**:545–550.
267. Deelen J, Evans DS, Arking DE, Tesi N, Nygaard M, Liu X, Wojczynski MK, Biggs ML, van der Spek A, Atzmon G, Ware EB, Sarnowski C, Smith AV, Seppälä I, Cordell HJ, Dose J, Amin N, Arnold AM, Ayers KL, Barzilai N, Becker EJ, Beekman M, Blanché H, Christensen K, Christiansen L, Collerton JC, Cubaynes S, Cummings SR, Davies K, Debrabant B, Deleuze JF, Duncan R, Faul JD, Franceschi C, Galan P, Gudnason V, Harris TB, Huisman M, Hurme MA, Jagger C, Jansen I, Jylhä M, Kähönen M, Karasik D, Kardia SLR, Kingston A, Kirkwood TBL, Launer LJ, Lehtimäki T, Lieb W, Lyytikäinen LP, Martin-Ruiz C, Min J, Nebel A, Newman AB, Nie C, Nohr EA, Orwoll ES, Perls TT, Province MA, Psaty BM, Raitakari OT, Reinders MJT, Robine JM, Rotter JI, Sebastiani P, Smith J, Sørensen TIA, Taylor KD, Uitterlinden AG, van der Flier W, van der Lee SJ, van Duijn CM, van Heemst D, Vaupel JW, Weir D, Ye K, Zeng Y, Zheng W, Holstege H, Kiel DP, Lunetta KL, Slagboom PE, Murabito JM. A meta-analysis of genome-wide association studies identifies multiple longevity genes. *Nat Commun* 2019;**10**:3669.
268. Sebastiani P, Gurinovich A, Nygaard M, Sasaki T, Sweigart B, Bae H, Andersen SL, Villa F, Atzmon G, Christensen K, Arai Y, Barzilai N, Puca A, Christiansen L, Hirose N, Perls TT. APOE alleles and extreme human longevity. *J Gerontol A Biol Sci Med Sci* 2019;**74**:44–51.
269. Snellman A, Ekblad LL, Tuisku J, Koivumäki M, Ashton NJ, Lantero-Rodriguez J, Karikari TK, Helin S, Bucci M, Löyttyniemi E, Parkkola R, Karrasch M, Schöll M, Zetterberg H, Blennow K, Rinne JO. APOE ϵ4 gene dose effect on imaging and blood biomarkers of neuroinflammation and beta-amyloid in cognitively unimpaired elderly. *Alzheimers Res Ther* 2023;**15**:71.
270. Di Battista AM, Heinsinger NM, Rebeck GW. Alzheimer's disease genetic risk factor APOE-ϵ4 also affects normal brain function. *Curr Alzheimer Res* 2016;**13**:1200–1207.
271. Mattace-Raso FU, van der Cammen TJ, van der Meer IM, Schalekamp MA, Asmar R, Hofman A, Witteman JC. C-reactive protein and arterial stiffness in older adults: the rotterdam study. *Atherosclerosis* 2004;**176**:111–116.
272. Desjardins MP, Sidibé A, Fortier C, Mac-Way F, Marquis K, De Serres S, Larivière R, Agharazii M. Association of interleukin-6 with aortic stiffness in end-stage renal disease. *J Am Soc Hypertens* 2018;**12**:5–13.
273. Blackburn R, Giral P, Bruckert E, André JM, Gonbert S, Bernard M, Chapman MJ, Turpin G. Elevated C-reactive protein constitutes an independent predictor of advanced carotid plaques in dyslipidemic subjects. *Arterioscler Thromb Vasc Biol* 2001;**21**:1962–1968.
274. Reilly MP, Wolfe ML, Localio AR, Rader DJ. C-reactive protein and coronary artery calcification. *Arterioscler Thromb Vasc Biol* 2003;**23**:1851–1856.
275. Libby P. Interleukin-1 beta as a target for atherosclerosis therapy: biological basis of CANTOS and beyond. *J Am Coll Cardiol* 2017;**70**:2278–2289.
276. Rosa M, Chignon A, Li Z, Boulanger MC, Arsenault BJ, Bossé Y, Thériault S, Mathieu P. A Mendelian randomization study of IL6 signaling in cardiovascular diseases, immune-related disorders and longevity. *NPJ Genom Med* 2019;**4**:23.
277. Interleukin 1 Genetics Consortium. Cardiometabolic effects of genetic upregulation of the interleukin 1 receptor antagonist: a Mendelian randomisation analysis. *Lancet Diabetes Endocrinol* 2015;**3**:243–253.
278. Arnett DK, Blumenthal RS, Albert MA, Buroker AB, Goldberger ZD, Hahn EJ, Himmelfarb CD, Khera A, Lloyd-Jones D, McEvoy JW, Michos ED, Miedema MD, Muñoz D, Smith SC Jr, Virani SS, Williams KA Sr, Yeboah J, Ziaeian B. 2019 ACC/AHA guideline on the primary prevention of cardiovascular disease: a report of the American College of Cardiology/American Heart Association task force on clinical practice guidelines. *Circulation* 2019; **140**:e596–e646.
279. Faghihi MA, Modarresi F, Khalil AM, Wood DE, Sahagan BG, Morgan TE, Finch CE, St Laurent G 3rd, Kenny PJ, Wahlestedt C. Expression of a noncoding RNA is elevated in Alzheimer's disease and drives rapid feed-forward regulation of beta-secretase. *Nat Med* 2008;**14**:723–730.
280. Li F, Wang Y, Yang H, Xu Y, Zhou X, Zhang X, Xie Z, Bi J. The effect of BACE1-AS on beta-amyloid generation by regulating BACE1 mRNA expression. *BMC Mol Biol* 2019; **20**:23.
281. Thomas T, Thomas G, McLendon C, Sutton T, Mullan M. beta-Amyloid-mediated vasoactivity and vascular endothelial damage. *Nature* 1996;**380**:168–171.
282. Jans DM, Martinet W, Van De Parre TJ, Herman AG, Bult H, Kockx MM, De Meyer GR. Processing of amyloid precursor protein as a biochemical link between atherosclerosis and Alzheimer's disease. *Cardiovasc Hematol Disord Drug Targets* 2006;**6**:21–34.
283. Tibolla G, Norata GD, Meda C, Arnaboldi L, Uboldi P, Piazza F, Ferrarese C, Corsini A, Maggi A, Vegeto E, Catapano AL. Increased atherosclerosis and vascular inflammation in APP transgenic mice with apolipoprotein E deficiency. *Atherosclerosis* 2010;**210**:78–87.
284. Van De Parre TJ, Guns PJ, Fransen P, Martinet W, Bult H, Herman AG, De Meyer GR. Attenuated atherogenesis in apolipoprotein E-deficient mice lacking amyloid precursor protein. *Atherosclerosis* 2011;**216**:54–58.
285. Mucchiano G, Cornwell GG 3rd, Westermark P. Senile aortic amyloid. Evidence for two distinct forms of localized deposits. *Am J Pathol* 1992;**140**:871–877.
286. Roher AE, Esh CL, Kokjohn TA, Castaño EM, Van Vickle GD, Kalback WM, Patton RL, Luehrs DC, Daugs ID, Kuo YM, Emmerling MR, Soares H, Quinn JF, Kaye J, Connor DJ, Silverberg NB, Adler CH, Seward JD, Beach TG, Sabbagh MN. Amyloid beta peptides in human plasma and tissues and their significance for Alzheimer's disease. *Alzheimers Dement* 2009;**5**:18–29.
287. De Meyer GR, De Cleen DM, Cooper S, Knaapen MW, Jans DM, Martinet W, Herman AG, Bult H, Kockx MM. Platelet phagocytosis and processing of beta-amyloid precursor protein as a mechanism of macrophage activation in atherosclerosis. *Circ Res* 2002;**90**:1197–1204.
288. Meakin PJ, Coull BM, Tuharska Z, McCaffery C, Akoumianakis I, Antoniades C, Brown J, Griffin KJ, Platt F, Ozber CH, Yuldasheva NY, Makava N, Skromna A, Prescott A, McNeilly AD, Siddiqui M, Palmer CN, Khan F, Ashford ML. Elevated circulating amyloid concentrations in obesity and diabetes promote vascular dysfunction. *J Clin Invest* 2020; **130**:4104–4117.
289. Tachida Y, Miura S, Muto Y, Takuwa H, Sahara N, Shindo A, Matsuba Y, Saito T, Taniguchi N, Kawaguchi Y, Tomimoto H, Saido T, Kitazume S. Endothelial expression of human amyloid precursor protein leads to amyloid beta in the blood and induces cerebral amyloid angiopathy in knock-in mice. *J Biol Chem* 2022;**298**:101880.
290. d'Uscio LV, Katusic ZS. Endothelium-specific deletion of amyloid-beta precursor protein exacerbates endothelial dysfunction induced by aging. *Aging (Albany NY)* 2021;**13**: 19165–19185.
291. Stamatelopoulos K, Pol CJ, Ayers C, Georgiopoulos G, Gatsiou A, Brilakis ES, Khera A, Drosatos K, de Lemos JA, Stellos K. Amyloid-Beta (1-40) peptide and subclinical cardiovascular disease. *J Am Coll Cardiol* 2018;**72**:1060–1061.
292. Zhu F, Wolters FJ, Yaqub A, Leening MJG, Ghanbari M, Boersma E, Ikram MA, Kavousi M. Plasma amyloid-beta in relation to cardiac function and risk of heart failure in general population. *JACC Heart Fail* 2023;**11**:93–102.
293. Janelidze S, Stomrud E, Palmqvist S, Zetterberg H, van Westen D, Jeromin A, Song L, Hanlon D, Tan Hehir CA, Baker D, Blennow K, Hansson O. Plasma beta-amyloid in Alzheimer's disease and vascular disease. *Sci Rep* 2016;**6**:26801.
294. Mavraganis G, Georgiopoulos G, Zervas G, Aivalioti E, Delialis D, Petropoulos I, Rachiotis N, Konstantaki C, Moustou C, Dimopoulou MA, Sachse M, Tual-Chalot S, Sopova K, Psimmenou E, Stellos K, Stamatelopoulos K. Circulating amyloid beta 1-40 peptide as an associate of renal function decline. *Eur J Clin Invest* 2025;**55**:e70006.
295. Lambrinoudaki I, Delialis D, Georgiopoulos G, Tual-Chalot S, Vlachogiannis NI, Patras R, Aivalioti E, Armeni E, Augoulea A, Tsoltos N, Soureti A, Stellos K, Stamatelopoulos K. Circulating amyloid Beta 1-40 is associated with increased rate of progression of atherosclerosis in menopause: a prospective cohort study. *Thromb Haemost* 2021;**121**:650–658.
296. Delialis D, Georgiopoulos G, Tual-Chalot S, Angelidakis L, Aivalioti E, Mavraganis G, Sopova K, Argyris A, Kostakou P, Konstantaki C, Papaioannou M, Tsilimigras D, Chatoupis K, Zacharoulis AA, Galyfos G, Sigala F, Stellos K, Stamatelopoulos K. Amyloid beta is associated with carotid wall echolucency and atherosclerotic plaque composition. *Sci Rep* 2024;**14**:14944.
297. Stamatelopoulos K, Sibbing D, Rallidis LS, Georgiopoulos G, Stakos D, Braun S, Gatsiou A, Sopova K, Kotakos C, Varounis C, Tellis CC, Kastritis E, Alevizaki M, Tselepis AD, Alexopoulos P, Laske C, Keller T, Kastrati A, Dimmeler S, Zeiher AM, Stellos K. Amyloid-beta (1-40) and the risk of death from cardiovascular causes in patients with coronary heart disease. *J Am Coll Cardiol* 2015;**65**:904–916.
298. Stamatelopoulos K, Mueller-Hennessen M, Georgiopoulos G, Sachse M, Boeddinghaus J, Sopova K, Gatsiou A, Amrhein C, Biener M, Vafaie M, Athanasouli F, Stakos D, Pateras K, Twerenbold R, Badertscher P, Nestelberger T, Dimmeler S, Katus HA, Zeiher AM, Mueller C, Giannitsis E, Stellos K. Amyloid-beta (1-40) and mortality in patients with non-ST-segment elevation acute coronary syndrome: a cohort study. *Ann Intern Med* 2018;**168**:855–865.
299. Dorsheimer L, Assmus B, Rasper T, Ortmann CA, Ecke A, Abou-El-Ardat K, Schmid T, Brüne B, Wagner S, Serve H, Hoffmann J, Seeger F, Dimmeler S, Zeiher AM, Rieger MA. Association of mutations contributing to clonal hematopoiesis with prognosis in chronic ischemic heart failure. *JAMA Cardiol* 2019;**4**:25–33.
300. Abplanalp WT, Cremer S, John D, Hoffmann J, Schuhmacher B, Merten M, Rieger MA, Vasa-Nicotera M, Zeiher AM, Dimmeler S. Clonal hematopoiesis-driver DNMT3A mutations Alter immune cells in heart failure. *Circ Res* 2021;**128**:216–228.
301. Jaiswal S, Natarajan P, Silver AJ, Gibson CJ, Bick AG, Shvartz E, McConkey M, Gupta N, Gabriel S, Ardissino D, Baber U, Mehran R, Fuster V, Danesh J, Frossard P, Saleheen D, Melander O, Sukhova GK, Neuberg D, Libby P, Kathiresan S, Ebert BL. Clonal hematopoiesis and risk of atherosclerotic cardiovascular disease. *N Engl J Med* 2017;**377**:111–121.
302. Pascual-Figal DA, Bayes-Genis A, Díez-Díez M, Hernández-Vicente Á, Vázquez-Andrés D, de la Barrera J, Vazquez E, Quintas A, Zuriaga MA, Asensio-López MC, Dopazo A, Sánchez-Cabo F, Fuster JJ. Clonal hematopoiesis and risk of progression of heart failure with reduced left ventricular ejection fraction. *J Am Coll Cardiol* 2021;**77**:1747–1759.
303. Mas-Peiro S, Hoffmann J, Fichtlscherer S, Dorsheimer L, Rieger MA, Dimmeler S, Vasa-Nicotera M, Zeiher AM. Clonal haematopoiesis in patients with degenerative aortic valve stenosis undergoing transcatheter aortic valve implantation. *Eur Heart J* 2020;**41**: 933–939.
304. Genovese G, Kähler AK, Handsaker RE, Lindberg J, Rose SA, Bakhoum SF, Chambert K, Mick E, Neale BM, Fromer M, Purcell SM, Svantesson O, Landén M, Höglund M, Lehmann S, Gabriel SB, Moran JL, Lander ES, Sullivan PF, Sklar P, Grönberg H, Hultman

CM, McCarroll SA. Clonal hematopoiesis and blood-cancer risk inferred from blood DNA sequence. *N Engl J Med* 2014;**371**:2477–2487.
305. Robertson NA, Latorre-Crespo E, Terradas-Terradas M, Lemos-Portela J, Purcell AC, Livesey BJ, Hillary RF, Murphy L, Fawkes A, MacGillivray L, Copland M, Marioni RE, Marsh JA, Harris SE, Cox SR, Deary IJ, Schumacher LJ, Kirschner K, Chandra T. Longitudinal dynamics of clonal hematopoiesis identifies gene-specific fitness effects. *Nat Med* 2022;**28**:1439–1446.
306. Heyde A, Rohde D, McAlpine CS, Zhang S, Hoyer FF, Gerold JM, Cheek D, Iwamoto Y, Schloss MJ, Vandoorne K, Iborra-Egea O, Muñoz-Guijosa C, Bayes-Genis A, Reiter JG, Craig M, Swirski FK, Nahrendorf M, Nowak MA, Naxerova K. Increased stem cell proliferation in atherosclerosis accelerates clonal hematopoiesis. *Cell* 2021;**184**:1348–1361.e22.
307. Pasupuleti SK, Ramdas B, Burns SS, Palam LR, Kanumuri R, Kumar R, Pandhiri TR, Dave UP, Yellapu NK, Zhou X, Zhang C, Sandusky GE, Yu Z, Honigberg MC, Bick AG, Griffin GK, Niroula A, Ebert BL, Paczesny S, Natarajan P, Kapur R. Obesity-induced inflammation exacerbates clonal hematopoiesis. *J Clin Invest* 2023;**133**:e163968.
308. Andersson-Assarsson JC, van Deuren RC, Kristensson FM, Steehouwer M, Sjöholm K, Svensson PA, Pieterse M, Gilissen C, Taube M, Jacobson P, Perkins R, Brunner HG, Netea MG, Peltonen M, Carlsson B, Hoischen A, Carlsson LMS. Evolution of age-related mutation-driven clonal haematopoiesis over 20 years is associated with metabolic dysfunction in obesity. *EBioMedicine* 2023;**92**:104621.
309. Kar SP, Quiros PM, Gu M, Jiang T, Mitchell J, Langdon R, Iyer V, Barcena C, Vijayabaskar MS, Fabre MA, Carter P, Petrovski S, Burgess S, Vassiliou GS. Genome-wide analyses of 200,453 individuals yield new insights into the causes and consequences of clonal hematopoiesis. *Nat Genet* 2022;**54**:1155–1166.
310. Rossiello F, Jurk D, Passos JF, d'Adda di Fagagna F. Telomere dysfunction in ageing and age-related diseases. *Nat Cell Biol* 2022;**24**:135–147.
311. Díez-Díez M, Ramos-Neble BL, de la Barrera J, Silla-Castro JC, Quintas A, Vázquez E, Rey-Martín MA, Izzi B, Sánchez-García L, García-Lunar I, Mendieta G, Mass V, Gómez-López N, Espadas C, González G, Quesada AJ, García-Álvarez A, Fernández-Ortiz A, Lara-Pezzi E, Dopazo A, Sánchez-Cabo F, Ibáñez B, Andrés V, Fuster V, Fuster JJ. Unidirectional association of clonal hematopoiesis with atherosclerosis development. *Nat Med* 2024;**30**:2857–2866.
312. Yalcinkaya M, Liu W, Thomas LA, Olszewska M, Xiao T, Abramowicz S, Papapetrou EP, Westerterp M, Wang N, Tabas I, Tall AR. BRCC3-Mediated NLRP3 deubiquitylation promotes inflammasome activation and atherosclerosis in Tet2 clonal hematopoiesis. *Circulation* 2023;**148**:1764–1777.
313. Redfield MM, Jacobsen SJ, Borlaug BA, Rodeheffer RJ, Kass DA. Age- and gender-related ventricular-vascular stiffening: a community-based study. *Circulation* 2005;**112**:2254–2262.
314. Robinson S, Ring L, Oxborough D, Harkness A, Bennett S, Rana B, Sutaria N, Lo Giudice F, Shun-Shin M, Paton M, Duncan R, Willis J, Colebourn C, Bassindale G, Gatenby K, Belham M, Cole G, Augustine D, Smiseth OA. The assessment of left ventricular diastolic function: guidance and recommendations from the British Society of Echocardiography. *Echo Res Pract* 2024;**11**:16.
315. Goette A, Corradi D, Dobrev D, Aguinaga L, Cabrera JA, Chugh SS, de Groot JR, Soulat-Dufour L, Fenelon G, Hatem SN, Jalife J, Lin YJ, Lip GYH, Marcus GM, Murray KT, Pak HN, Schotten U, Takahashi N, Yamaguchi T, Zoghbi WA, Nattel S, Mont L, Akar JG, Akoum N, Althoff T, Diaz JC, Guichard JB, Jadidi A, Kalman J, Lim H, Teixeira RA. Atrial cardiomyopathy revisited—evolution of a concept: a clinical consensus statement of the European Heart Rhythm Association (EHRA) of the ESC, the Heart Rhythm Society (HRS), the Asian Pacific Heart Rhythm Society (APHRS), and the Latin American Heart Rhythm Society (LAHRS). *EP Europace* 2024;**26**:euae204.
316. Kamel H, Longstreth WT Jr, Tirschwell DL, Kronmal RA, Marshall RS, Broderick JP, Aragón García R, Plummer P, Sabagha N, Pauls Q, Cassarly C, Dillon CR, Di Tullio MR, Hod EA, Soliman EZ, Gladstone DJ, Healey JS, Sharma M, Chaturvedi S, Janis LS, Krishnaiah B, Nahab F, Kasner SE, Stanton RJ, Kleindorfer DO, Starr M, Winder TR, Clark WM, Miller BR, Elkind MSV; ARCADIA Investigators. Apixaban to prevent recurrence after cryptogenic stroke in patients with atrial cardiopathy: the ARCADIA randomized clinical trial. *JAMA* 2024;**331**:573–581.
317. Touboul PJ, Hennerici MG, Meairs S, Adams H, Amarenco P, Bornstein N, Csiba L, Desvarieux M, Ebrahim S, Hernandez Hernandez R, Jaff M, Kownator S, Naqvi T, Prati P, Rundek T, Sitzer M, Schminke U, Tardif JC, Taylor A, Vicaut E, Woo KS. Mannheim carotid intima-media thickness and plaque consensus (2004–2006–2011). an update on behalf of the advisory board of the 3rd, 4th and 5th watching the risk symposia, at the 13th, 15th and 20th European Stroke Conferences, Mannheim, Germany, 2004, Brussels, Belgium, 2006, and Hamburg, Germany, 2011. *Cerebrovasc Dis* 2012;**34**:290–296.
318. Visseren FLJ, Mach F, Smulders YM, Carballo D, Koskinas KC, Bäck M, Benetos A, Biffi A, Boavida JM, Capodanno D, Cosyns B, Crawford C, Davos CH, Desormais I, Di Angelantonio E, Franco OH, Halvorsen S, Hobbs FDR, Hollander M, Jankowska EA, Michal M, Sacco S, Sattar N, Tokgozoglu L, Tonstad S, Tsioufis KP, van Dis I, van Gelder IC, Wanner C, Williams B; ESC National Cardiac Societies; ESC Scientific Document Group. 2021 ESC Guidelines on cardiovascular disease prevention in clinical practice: developed by the Task Force for cardiovascular disease prevention in clinical practice with representatives of the European Society of Cardiology and 12 medical societies with the special contribution of the European Association of Preventive Cardiology (EAPC). *Eur Heart J* 2021;**42**:3227–3337.
319. Ge J, Jing F, Ji R, Tian A, Su X, Li W, He G, Pu B, Lei L, Lu J, Li J. Age-related trends in the predictive value of carotid intima-Media thickness for cardiovascular death: a prospective population-based cohort study. *J Am Heart Assoc* 2023;**12**:e029656.
320. Zhou J, Li C, Cong H, Duan L, Wang H, Wang C, Tan Y, Liu Y, Zhang Y, Zhou X, Zhang H, Wang X, Ma Y, Yang J, Chen Y, Guo Z. Comparison of different investigation strategies to defer cardiac testing in patients with stable chest pain. *JACC Cardiovasc Imaging* 2022;**15**:91–104.
321. Newman AB, Naydeck BL, Sutton-Tyrrell K, Feldman A, Edmundowicz D, Kuller LH. Coronary artery calcification in older adults to age 99. *Circulation* 2001;**104**:2679–2684.
322. Vrints C, Andreotti F, Koskinas KC, Rossello X, Adamo M, Ainslie J, Banning AP, Budaj A, Buechel RR, Chiariello GA, Chieffo A, Christodorescu RM, Deaton C, Doenst T, Jones HW, Kunadian V, Mehilli J, Milojevic M, Piek JJ, Pugliese F, Rubboli A, Semb AG, Senior R, Ten Berg JM, Van Belle E, Van Craenenbroeck EM, Vidal-Perez R, Winther S; ESC Scientific Document Group. 2024 ESC Guidelines for the management of chronic coronary syndromes: developed by the task force for the management of chronic coronary syndromes of the European Society of Cardiology (ESC) endorsed by the European Association for Cardio-Thoracic Surgery (EACTS). *Eur Heart J* 2024;**45**:3415–3537.
323. Blaha MJ, Naazie IN, Cainzos-Achirica M, Dardari ZA, DeFilippis AP, McClelland RL, Mirbolouk M, Orimoloye OA, Dzaye O, Nasir K, Page JH. Derivation of a coronary age calculator using traditional risk factors and coronary artery calcium: the multi-ethnic study of atherosclerosis. *J Am Heart Assoc* 2021;**10**:e019351.
324. Ferreira VM, Schulz-Menger J, Holmvang G, Kramer CM, Carbone I, Sechtem U, Kindermann I, Gutberlet M, Cooper LT, Liu P, Friedrich MG. Cardiovascular magnetic resonance in nonischemic myocardial inflammation: expert recommendations. *J Am Coll Cardiol* 2018;**72**:3158–3176.
325. O'Brien AT, Gil KE, Varghese J, Simonetti OP, Zareba KM. T2 mapping in myocardial disease: a comprehensive review. *J Cardiovasc Magn Reson* 2022;**24**:33.
326. Shah M, de A Inácio MH, Lu C, Schiratti PR, Zheng SL, Clement A, de Marvao A, Bai W, King AP, Ware JS, Wilkins MR, Mielke J, Elci E, Kryukov I, McGurk KA, Bender C, Freitag DF, O'Regan DP. Environmental and genetic predictors of human cardiovascular ageing. *Nat Commun* 2023;**14**:4941.
327. Bai W, Suzuki H, Huang J, Francis C, Wang S, Tarroni G, Guitton F, Aung N, Fung K, Petersen SE, Piechnik SK, Neubauer S, Evangelou E, Dehghan A, O'Regan DP, Wilkins MR, Guo Y, Matthews PM, Rueckert D. A population-based phenome-wide association study of cardiac and aortic structure and function. *Nat Med* 2020;**26**:1654–1662.
328. Thanaj M, Mielke J, McGurk KA, Bai W, Savioli N, de Marvao A, Meyer HV, Zeng L, Sohler F, Lumbers RT, Wilkins MR, Ware JS, Bender C, Rueckert D, MacNamara A, Freitag DF, O'Regan DP. Genetic and environmental determinants of diastolic heart function. *Nat Cardiovasc Res* 2022;**1**:361–371.
329. Ajoolabady A, Pratico D, Ren J. Endothelial dysfunction: mechanisms and contribution to diseases. *Acta Pharmacol Sin* 2024;**45**:2023–2031.
330. Reference Values for Arterial Stiffness' Collaboration. Determinants of pulse wave velocity in healthy people and in the presence of cardiovascular risk factors: 'establishing normal and reference values'. *Eur Heart J* 2010;**31**:2338–2350.
331. Blacher J, Guerin AP, Pannier B, Marchais SJ, Safar ME, London GM. Impact of aortic stiffness on survival in end-stage renal disease. *Circulation* 1999;**99**:2434–2439.
332. Choi CU, Park EB, Suh SY, Kim JW, Kim EJ, Rha SW, Seo HS, Oh DJ, Park CG. Impact of aortic stiffness on cardiovascular disease in patients with chest pain: assessment with direct intra-arterial measurement. *Am J Hypertens* 2007;**20**:1163–1169.
333. Cruickshank K, Riste L, Anderson SG, Wright JS, Dunn G, Gosling RG. Aortic pulse-wave velocity and its relationship to mortality in diabetes and glucose intolerance: an integrated index of vascular function? *Circulation* 2002;**106**:2085–2090.
334. Shokawa T, Imazu M, Yamamoto H, Toyofuku M, Tasaki N, Okimoto T, Yamane K, Kohno N. Pulse wave velocity predicts cardiovascular mortality: findings from the Hawaii-Los Angeles-Hiroshima study. *Circ J* 2005;**69**:259–264.
335. Willum-Hansen T, Staessen JA, Torp-Pedersen C, Rasmussen S, Thijs L, Ibsen H, Jeppesen J. Prognostic value of aortic pulse wave velocity as index of arterial stiffness in the general population. *Circulation* 2006;**113**:664–670.
336. Marshall AG, Neikirk K, Afolabi J, Mwesigwa N, Shao B, Kirabo A, Reddy AK, Hinton A Jr. Update on the use of pulse wave velocity to measure age-related vascular changes. *Curr Hypertens Rep* 2024;**26**:131–140.
337. Ludmer PL, Selwyn AP, Shook TL, Wayne RR, Mudge GH, Alexander RW, Ganz P. Paradoxical vasoconstriction induced by acetylcholine in atherosclerotic coronary arteries. *N Engl J Med* 1986;**315**:1046–1051.
338. Thijssen DHJ, Bruno RM, van Mil ACCM, Holder SM, Faita F, Greyling A, Zock PL, Taddei S, Deanfield JE, Luscher T, Green DJ, Ghiadoni L. Expert consensus and evidence-based recommendations for the assessment of flow-mediated dilation in humans. *Eur Heart J* 2019;**40**:2534–2547.
339. Charakida M, Masi S, Lüscher TF, Kastelein JJ, Deanfield JE. Assessment of atherosclerosis: the role of flow-mediated dilatation. *Eur Heart J* 2010;**31**:2854–2861.
340. Rossi R, Nuzzo A, Olaru AI, Origliani G, Modena MG. Endothelial function affects early carotid atherosclerosis progression in hypertensive postmenopausal women. *J Hypertens* 2011;**29**:1136–1144.
341. Inaba Y, Chen JA, Bergmann SR. Prediction of future cardiovascular outcomes by flow-mediated vasodilatation of brachial artery: a meta-analysis. *Int J Cardiovasc Imaging* 2010;**26**:631–640.
342. Ras RT, Streppel MT, Draijer R, Zock PL. Flow-mediated dilation and cardiovascular risk prediction: a systematic review with meta-analysis. *Int J Cardiol* 2013;**168**:344–351.
343. Matsuzawa Y, Kwon TG, Lennon RJ, Lerman LO, Lerman A. Prognostic value of flow-mediated vasodilation in brachial artery and fingertip artery for cardiovascular events: a systematic review and meta-analysis. *J Am Heart Assoc* 2015;**4**:e002270.

344. Axtell AL, Gomari FA, Cooke JP. Assessing endothelial vasodilator function with the Endo-PAT 2000. *J Vis Exp* 2010;**44**:2167.
345. Bonetti PO, Pumper GM, Higano ST, Holmes DR, Kuvin JT, Lerman A. Noninvasive identification of patients with early coronary atherosclerosis by assessment of digital reactive hyperemia. *J Am Coll Cardiol* 2004;**44**:2137–2141.
346. Rubinshtein R, Kuvin JT, Soffler M, Lennon RJ, Lavi S, Nelson RE, Pumper GM, Lerman LO, Lerman A. Assessment of endothelial function by non-invasive peripheral arterial tonometry predicts late cardiovascular adverse events. *Eur Heart J* 2010;**31**:1142–1148.
347. D'Agostino RB Sr, Grundy S, Sullivan LM, Wilson P. Validation of the Framingham coronary heart disease prediction scores: results of a multiple ethnic groups investigation. *JAMA* 2001;**286**:180–187.
348. Aktas MK, Ozduran V, Pothier CE, Lang R, Lauer MS. Global risk scores and exercise testing for predicting all-cause mortality in a preventive medicine program. *JAMA* 2004;**292**:1462–1468.
349. Hippisley-Cox J, Coupland CAC, Bafadhel M, Russell REK, Sheikh A, Brindle P, Channon KM. Development and validation of a new algorithm for improved cardiovascular risk prediction. *Nat Med* 2024;**30**:1440–1447.
350. Wang RS, Maron BA, Loscalzo J. Multiomics network medicine approaches to precision medicine and therapeutics in cardiovascular diseases. *Arterioscler Thromb Vasc Biol* 2023;**43**:493–503.
351. Oh HS, Rutledge J, Nachun D, Pálovics R, Abiose O, Moran-Losada P, Channappa D, Urey DY, Kim K, Sung YJ, Wang L, Timsina J, Western D, Liu M, Kohlfeld P, Budde J, Wilson EN, Guen Y, Maurer TM, Haney M, Yang AC, He Z, Greicius MD, Andreasson KI, Sathyan S, Weiss EF, Milman S, Barzilai N, Cruchaga C, Wagner AD, Mormino E, Lehallier B, Henderson VW, Longo FM, Montgomery SB, Wyss-Coray T. Organ aging signatures in the plasma proteome track health and disease. *Nature* 2023;**624**:164–172.
352. Kivimäki M, Frank P, Pentti J, Jokela M, Nyberg ST, Blake A, Lindbohm JV, Oh HS, Singh-Manoux A, Wyss-Coray T, Partridge L. Proteomic organ-specific ageing signatures and 20-year risk of age-related diseases: the Whitehall II observational cohort study. *Lancet Digit Health* 2025;**7**:e195–e204.
353. Dorraki M, Liao Z, Abbott D, Psaltis PJ, Baker E, Bidargaddi N, Wardill HR, van den Hengel A, Narula J, Verjans JW. Improving cardiovascular disease prediction with machine learning using mental health data: a prospective UK Biobank study. *JACC Adv* 2024;**3**:101180.
354. Moqri M, Herzog C, Poganik JR; Biomarkers of Aging Consortium; Justice J, Belsky DW, Higgins-Chen A, Moskalev A, Fuellen G, Cohen AA, Bautmans I, Widschwendter M, Ding J, Fleming A, Mannick J, Han JJ, Zhavoronkov A, Barzilai N, Kaeberlein M, Cummings S, Kennedy BK, Ferrucci L, Horvath S, Verdin E, Maier AB, Snyder MP, Sebastiano V, Gladyshev VN. Biomarkers of aging for the identification and evaluation of longevity interventions. *Cell* 2023;**186**:3758–3775.
355. Mamoshina P, Kochetov K, Putin E, Cortese F, Aliper A, Lee WS, Ahn SM, Uhn L, Skjodt N, Kovalchuk O, Scheibye-Knudsen M, Zhavoronkov A. Population specific biomarkers of human aging: a big data study using South Korean, Canadian, and Eastern European patient populations. *J Gerontol A Biol Sci Med Sci* 2018;**73**:1482–1490.
356. Macdonald-Dunlop E, Taba N, Klarić L, Frkatović A, Walker R, Hayward C, Esko T, Haley C, Fischer K, Wilson JF, Joshi PK. A catalogue of omics biological ageing clocks reveals substantial commonality and associations with disease risk. *Aging (Albany NY)* 2022;**14**:623–659.
357. Ganz P, Heidecker B, Hveem K, Jonasson C, Kato S, Segal MR, Sterling DG, Williams SA. Development and validation of a protein-based risk score for cardiovascular outcomes among patients with stable coronary heart disease. *JAMA* 2016;**315**:2532–2541.
358. Aging Biomarker Consortium; Bao H, Cao J, Chen M, Chen M, Chen W, Chen X, Chen Y, Chen Y, Chen Y, Chen Z, Chhetri JK, Ding Y, Feng J, Guo J, Guo M, He C, Jia Y, Jiang H, Jing Y, Li D, Li J, Li J, Liang Q, Liang R, Liu F, Liu X, Liu Z, Luo OJ, Lv J, Ma J, Mao K, Nie J, Qiao X, Sun X, Tang X, Wang J, Wang Q, Wang S, Wang X, Wang Y, Wang Y, Wu R, Xia K, Xiao FH, Xu L, Xu Y, Yan H, Yang L, Yang R, Yang Y, Ying Y, Zhang L, Zhang W, Zhang W, Zhang X, Zhang Z, Zhou M, Zhou R, Zhu Q, Zhu Z, Cao F, Cao Z, Chan P, Chen C, Chen G, Chen HZ, Chen J, Ci W, Ding BS, Ding Q, Gao F, Han JJ, Huang K, Ju Z, Kong QP, Li J, Li J, Li X, Liu B, Liu F, Liu L, Liu Q, Liu Q, Liu X, Liu Y, Luo X, Ma S, Ma X, Mao Z, Nie J, Peng Y, Qu J, Ren J, Ren R, Song M, Songyang Z, Sun YE, Sun Y, Tian M, Wang S, Wang S, Wang X, Wang X, Wang YJ, Wang Y, Wong CCL, Xiang AP, Xiao Y, Xie Z, Xu D, Ye J, Yue R, Zhang C, Zhang H, Zhang L, Zhang W, Zhang Y, Zhang YW, Zhang Z, Zhao T, Zhao Y, Zhu D, Zou W, Pei G, Liu GH. Biomarkers of aging. *Sci China Life Sci* 2023;**66**:893–1066.
359. Sayed N, Huang Y, Nguyen K, Krejciova-Rajaniemi Z, Grawe AP, Gao T, Tibshirani R, Hastie T, Alpert A, Cui L, Kuznetsova T, Rosenberg-Hasson Y, Ostan R, Monti D, Lehallier B, Shen-Orr SS, Maecker HT, Dekker CL, Wyss-Coray T, Franceschi C, Jojic V, Haddad F, Montoya JG, Wu JC, Davis MM, Furman D. An inflammatory aging clock (iAge) based on deep learning tracks multimorbidity, immunosenescence, frailty and cardiovascular aging. *Nat Aging* 2021;**1**:598–615.
360. Alpert A, Pickman Y, Leipold M, Rosenberg-Hasson Y, Ji X, Gaujoux R, Rabani H, Starosvetsky E, Kveler K, Schaffert S, Furman D, Caspi O, Rosenschein U, Khatri P, Dekker CL, Maecker HT, Davis MM, Shen-Orr SS. A clinically meaningful metric of immune age derived from high-dimensional longitudinal monitoring. *Nat Med* 2019;**25**:487–495.
361. Kabacik S, Lowe D, Fransen L, Leonard M, Ang SL, Whiteman C, Corsi S, Cohen H, Felton S, Bali R, Horvath S, Raj K. The relationship between epigenetic age and the hallmarks of aging in human cells. *Nat Aging* 2022;**2**:484–493.
362. Mitchell GF. Arterial stiffness in aging: does it have a place in clinical practice? : recent advances in hypertension. *Hypertension* 2021;**77**:768–780.
363. Chen C, Ding S, Wang J. Digital health for aging populations. *Nat Med* 2023;**29**:1623–1630.
364. Samargandy S, Matthews KA, Brooks MM, Barinas-Mitchell E, Magnani JW, Janssen I, Hollenberg SM, El Khoudary SR. Arterial stiffness accelerates within 1 year of the final menstrual period: the SWAN heart study. *Arterioscler Thromb Vasc Biol* 2020;**40**:1001–1008.
365. Bruno RM, Nilsson PM, Engström G, Wadström BN, Empana JP, Boutouyrie P, Laurent S. Early and supernormal vascular aging: clinical characteristics and association with incident cardiovascular events. *Hypertension* 2020;**76**:1616–1624.
366. O'Kelly AC, Michos ED, Shufelt CL, Vermunt JV, Minissian MB, Quesada O, Smith GN, Rich-Edwards JW, Garovic VD, El Khoudary SR, Honigberg MC. Pregnancy and reproductive risk factors for cardiovascular disease in women. *Circ Res* 2022;**130**:652–672.
367. Zink F, Stacey SN, Norddahl GL, Frigge ML, Magnusson OT, Jonsdottir I, Thorgeirsson TE, Sigurdsson A, Gudjonsson SA, Gudmundsson J, Jonasson JG, Tryggvadottir L, Jonsson T, Helgason A, Gylfason A, Sulem P, Rafnar T, Thorsteinsdottir U, Gudbjartsson DF, Masson G, Kong A, Stefansson K. Clonal hematopoiesis, with and without candidate driver mutations, is common in the elderly. *Blood* 2017;**130**:742–752.
368. Schuermans A, Honigberg MC. Clonal haematopoiesis in cardiovascular disease: prognostic role and novel therapeutic target. *Nat Rev Cardiol* 2025.