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# AI-driven biological clocks of cardiac aging: Should we integrate ECG, metabolomic, and epigenetic signatures?

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**Systematic Review**

**Keywords:** Biological aging clocks, artificial intelligence, electrocardiogram, metabolomics, epigenetics, cardiovascular disease, gut microbiome, TMAO, multi-omics integration, personalized medicine

**Posted Date:** July 23rd, 2026

**DOI:** https://doi.org/10.21203/rs.3.rs-10155822/v1

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**Additional Declarations:** No competing interests reported.

# Abstract

### Background

Cardiovascular diseases remain the leading cause of death globally, with age as the primary risk factor. Unlike chronological age, biological age aims to capture age-related physiological changes and may better predict health outcomes and disease risk. Recent advances in artificial intelligence have enabled the development of aging clocks, computational models that estimate biological age from diverse biomarker patterns.

### Main body

This review examines three complementary approaches to quantifying cardiac and systemic aging. Epigenetic clocks, particularly second-generation models such as GrimAge, currently represent the most established molecular measures of biological aging and show strong associations with cardiovascular outcomes. AI-based electrocardiogram (ECG) clocks capture functional cardiac aging, with landmark studies demonstrating that ECG age gaps independently predict mortality and that accelerated ECG aging may confer up to 79% higher mortality risk. Metabolomic clocks provide complementary insight into systemic metabolic aging and reveal links between metabolic dysregulation and cardiovascular aging, including associations involving metabolites such as trimethylamine N-oxide (TMAO). However, current aging clocks face important limitations: single-modality approaches capture only one dimension of aging, the biological mechanisms underlying accelerated aging remain incompletely understood, and population generalizability remains limited.

### Conclusions

Integration of ECG, metabolomic, and epigenetic data promises a paradigm shift from reactive disease management to proactive, personalized aging intervention. Understanding concordance and discordance between these complementary clocks may uncover distinct aging phenotypes and reveal modifiable biological pathways. Future research should focus on integrating multi-omics data within large longitudinal cohorts to build interpretable, population-generalizable models that can guide clinical interventions to extend healthspan and prevent cardiovascular disease.


# Background

Cardiovascular disease (CVD) is the leading cause of death in the world, accounting for approximately 20.5 million deaths in 2025 and projected to rise substantially in the coming decades [1]. Its prevention and treatment are therefore crucial. Aging is the leading risk factor for CVD and represents a complex, multifaceted process. Contrary to chronological age, which measures the years since birth, biological age aims to capture an individual's physiological and molecular state and may better reflect heterogeneity in aging trajectories. Genetic, environmental, and lifestyle factors, including diet, stress levels, exercise, and sleep quality, can influence biological aging and contribute to differences in health

outcomes among individuals of the same chronological age [2]. Recent advances in multi-omic and clinical data integration have enabled the estimation of biological age using multiple data types, such as methylation, imaging, gene expression, and microbiome. These predictions can then be associated with disease risk, general well-being, and mortality [3].

## The challenge: understanding accelerated cardiac aging driven by cardiometabolic disease

Cardiometabolic diseases, including diabetes, obesity, and metabolic syndrome, are major systemic risk factors for cardiovascular disease and are associated with accelerated biological and cardiac aging. Chronic respiratory diseases, particularly COPD, may also contribute to accelerated cardiovascular aging through shared inflammatory and systemic mechanisms [4]. However, chronological age poorly predicts individual cardiovascular risk. Current single-modality approaches capture only part of the aging signatures, and the mechanistic links between systemic metabolism (especially microbial metabolites), molecular aging, and cardiac function remain unclear. Developing integrated multimodal aging clocks could identify individuals aging faster and reveal the metabolic, microbial, and epigenetic mechanisms driving accelerated cardiac aging. Figure 1 illustrates the relationship between chronological and biological age: deviations from the normal aging trajectory indicate accelerated or decelerated aging.

Computational models known as aging clocks have been developed to estimate biological age and aging rate using biomarker patterns across multiple biological layers, including proteomic, epigenetic, immunologic, and microbial markers. These estimates are associated with mortality risk and age-related health outcomes [5, 6]. In recent years, artificial intelligence (AI) has become an important tool in aging research for integrating and analyzing various omics types, including transcriptomics, proteomics, and epigenomics [3]. Deep learning (DL) approaches have shown promise for capturing complex nonlinear relationships in large, high-dimensional datasets, although their performance relative to conventional machine learning (ML) methods remains dependent on factors such as dataset size, data quality, and prediction task [7]. This review examines data-driven approaches to quantify cardiac and systemic aging through three complementary biological layers: AI-based ECG clocks that capture functional cardiac aging, metabolomic clocks that reflect systemic metabolic aging, and epigenetic clocks that quantify molecular aging. Integrating these distinct perspectives may improve understanding of the mechanisms driving accelerated cardiac aging and help identify individuals exhibiting faster biological aging. Figure 2 illustrates the conceptual framework for integrating ECG, metabolomic, and epigenetic aging clocks to study cardiac aging across complementary biological layers.

Cardiac aging & ECG-based clocks

The ECG is one of the most widely used clinical tools for assessing cardiac electrical activity and screening for cardiovascular disease [8]. Beyond its traditional role in diagnosing arrhythmias and structural abnormalities, the ECG provides a non-invasive window into the cumulative effects of aging on cardiac structure and function [9]. Detectable anomalies in the ECG include, for example, rhythm and

conduction disturbances and abnormal signal morphology [10]. However, aging manifests on the ECG through more subtle changes that extend beyond obvious pathology, making it an ideal candidate for biological cardiac age estimation.

Physiological basis: how aging affects the ECG

Cardiac aging causes two main adaptations visible on an ECG: fibrosis and ventricular hypertrophy [9]. These cause measurable changes in the electrical activity of the heart and ECG parameters, including P-wave duration, PR interval, QRS duration, and QTc interval, all of which increase significantly with age. Beyond structural changes, aging affects the cardiac autonomic nervous system, leading to changes in heart rate variability and circadian rhythms [10]. Such age-related changes accumulate gradually over decades and reflect intrinsic cardiac aging as well as CVD-related structural and electrical remodeling. ECG thus constitutes a dynamic marker that integrates multiple dimensions of cardiac aging.

As the heart is continuously active, highly energy-dependent, and affected by nearly all major aging processes (metabolic, cellular, and vascular), even small systemic changes, such as inflammation, oxidative stress, and mitochondrial decline, leave measurable traces in cardiac structure and function. These characteristics make the cardiovascular system particularly informative for studying aging, as one can assess age-related functional changes non-invasively and longitudinally through widely available tools such as ECGs. Unlike individual molecular biomarkers that capture specific biological processes, ECG-derived measures capture the integrated functional state of the cardiovascular system and can be monitored longitudinally through repeated non-invasive assessments. Cardiac aging also responds to lifestyle and pharmacological interventions, making it a potentially dynamic biomarker.

## Artificial intelligence for ECG age prediction

Although statistical models had been used to predict cardiac age from ECG features [5], Attia *et al*. (2019) pioneered the use of AI by training a convolutional neural network (CNN) on 499,727 12-lead ECGs to predict both age and sex [8]. The 12-lead ECG is the standard clinical electrocardiographic recording method, providing multiple electrical views of the heart from different anatomical orientations. Their model achieved 90.4% accuracy for sex classification and predicted age as a continuous variable with a mean absolute error (MAE) of 6.9 ± 5.6 years ($R^2 = 0.7$). Critically, in patients where CNN-predicted age exceeded chronological age by more than 7 years, there were significantly higher rates of low ejection fraction, hypertension, and coronary disease ($P < 0.01$), suggesting that the “ECG age gap” captures underlying cardiovascular damage not reflected in chronological age alone.

The landmark CODE study: deep neural network-estimated ECG age

A pivotal advance came with a deep CNN with residual functions (ResNet) trained on the massive CODE (Clinical Outcomes in Digital Electrocardiography) cohort comprising 1,558,415 patients from the Telehealth Network of Minas Gerais, Brazil [6]. This cohort included ECGs collected from 811 counties in the state of Minas Gerais between 2010 and 2017, one of the largest ECG datasets ever assembled for

aging research. The authors trained the model to predict chronological age from raw 12-lead ECG signals using a residual neural network architecture.

In a 15% hold-out test set (CODE-15%), patients with an ECG age over 8 years greater than their chronological age demonstrated significantly higher mortality rates (hazard ratio [HR] 1.79, $p < 0.001$). In contrast, those with an ECG age over 8 years below their chronological age showed reduced mortality (HR 0.78, $p < 0.001$). These findings were independently replicated in two external validation cohorts: ELSA-Brasil (Brazilian Longitudinal Study of Adult Health, *n* = 14,236 civil servants), with similar associations for both accelerated and decelerated ECG age (HR 1.75, 95% CI 1.35–2.27 and HR 0.74, 95% CI 0.63–0.88, respectively). In SaMi-Trop (São Paulo-Minas Gerais Tropical Medicine Research Center, *n* = 1,631 patients with chronic Chagas cardiomyopathy), where accelerated ECG age remained strongly associated with mortality (HR 2.42, 95% CI 1.53–3.83), whereas the association for decelerated ECG age was not statistically significant (HR 0.89, 95% CI 0.52–1.54, $p = 0.68$). Together, these results demonstrated robust generalizability across populations ranging from healthy adults to patients with advanced cardiac disease.

Perhaps most remarkably, even among patients with apparently normal ECGs by conventional diagnostic criteria, the predicted ECG age gap from chronological age remained a statistically significant predictor of mortality risk. This finding suggests that AI models can detect subtle aging-related changes in cardiac electrical activity that are imperceptible to both human experts and traditional automated ECG interpretation algorithms.

Advanced risk stratification: the AIRE platform

More recently, Fu Siong Ng and colleagues at Imperial College London developed the AI-ECG Risk Estimation (AIRE) platform, which uses DL combined with a discrete-time survival model to create patient-specific survival curves from a single ECG [11]. AIRE not only predicts mortality risk but also time-to-mortality, achieving C-indices (a measure of model discrimination indicating how well predicted risks correctly rank individuals by outcome, where 0.5 = random chance, 1.0 = perfect prediction) ranging from 0.638 to 0.775 across several validation cohorts from the USA, UK, and Brazil. To explore the biological underpinnings, genome-wide association analyses were performed using AIRE-predicted risk as a quantitative phenotype. This approach identified four genome-wide significant loci adjacent to *TBX3*, *VGLL2*, *CCDC91*, and *KCNQ1* which have been linked to cardiac structure and aging, including determinants of left ventricular mass and metabolic syndrome-related traits, suggesting that AI-derived ECG risk captures heritable aspects of cardiovascular remodeling and systemic aging [11].

Clinical relevance of ECG gap

The discrepancy between chronological age and ECG-predicted biological age, referred to as the ECG age gap, is typically calculated as the difference between predicted ECG age and chronological age (ECG age – chronological age) [9], although some studies incorporate age-adjusted residualization approaches, in which the age gap is regressed on chronological age to reduce age-dependent bias in

downstream analyses. The ECG age gap has proven to be a valuable biomarker of cardiovascular health. A positive ECG age gap indicates accelerated biological aging relative to chronological age, whereas a negative gap reflects slower-than-expected aging. Longitudinal studies using serial ECGs from the same individuals reveal that changes in ECG-predicted biological age over time are increasingly associated with cardiovascular risk, with hazard ratios for mortality increasing from 1.43 for single ECGs to 1.65 for longitudinal measurements. Furthermore, greater ECG aging effects are associated with higher odds ratios for atrial fibrillation, heart failure, and mortality [9].

The ECG age gap is modifiable by lifestyle factors: in a large UK Biobank analysis of 44,094 individuals, the presence of four favorable lifestyle factors (healthy diet, low alcohol consumption, high physical activity, and no smoking) was associated with an average of 2.5 years younger predicted ECG age than those with an unfavorable lifestyle [12]. This suggests that the ECG age gap not only identifies accelerated aging but may also be responsive to interventions, potentially making it a possible tool for monitoring biological age reversibility.

Beyond mortality prediction, AI-ECG models have demonstrated utility in detecting structural cardiac abnormalities: in patients with echocardiographic left ventricular ejection fraction (LVEF) ≥ 50%, those with AI-predicted LVEF ≤ 40% (“false positives”) actually had significantly higher rates of major adverse cardiovascular events (27.2% vs. 11.9%, $P < 0.001$) and mortality (20.4% vs. 9.6%, $P < 0.001$), suggesting these “discordant” predictions may identify subclinical disease not captured by conventional imaging [13].

Model architectures and interpretability

Most ECG age-prediction models rely on one-dimensional CNNs to extract temporal features from raw 12-lead signals. For example, the CODE study applied a ResNet specifically adapted to one-dimensional ECG signals, using multiple residual blocks to learn patterns in the waveform efficiently, and achieved robust performance across different populations [6]. Attia *et al.*’s model used a series of layers designed to first capture temporal patterns within each ECG lead and then combine information across leads, allowing it to detect both time-series dynamics and inter-lead relationships [8]. An overview of key ECG-based aging models, their training cohorts, performance metrics, and validation status is summarized in Table 1.

One major problem when applying ML models in medicine is that they are often treated as “black boxes”, making it difficult to understand how they reach their conclusions, despite high accuracy. The difficulty in understanding the reasoning behind a model’s output often makes clinicians hesitant to use or trust them [14]. However, the “black box” can often be, at least partially, opened with explainable AI tools. For instance, using the *post-hoc attribution method integrated gradients*, authors showed that models estimating a person’s ECG age focus on the same ECG characteristics that change naturally with chronological age, such as P-wave duration, PR interval, QRS duration, and QTc prolongation. In other words, the model’s pattern recognition aligns with what clinicians already know about how ECGs evolve

with age, and this alignment between model-derived patterns and clinical knowledge supports these predictions [9].

However, despite progress in explainability, significant challenges remain. As Lima *et al.* acknowledged, while exploratory analyses, including sensitivity analysis and expert ECG review, provide some insights into what the models detect, "it is far from sufficient to completely explain the findings" [6]. More fundamentally, the exact mechanisms by which AI models infer biological age from surface ECGs and which specific biological processes (such as metabolic dysfunction, inflammation, or microbiome-mediated pathways) contribute to the detectable ECG changes remain largely unknown.

**Table 1. Major AI-based ECG aging studies.**

Summary of landmark artificial intelligence-based electrocardiogram aging clock studies.

| Study | Cohort | Sample Size | Population | AI Method | Performance | Key Findings | External Validation |
|---|---|---|---|---|---|---|---|
| Attia 2019 | Mayo Clinic | 774 783 | Mixed clinical population (18-90 years) | 1D CNN | MAE 6.9 ± 5.6 years $R^2$ = 0.7 | ECG age gap > 7 years: higher CVD risk | No |
| Lima 2021 | CODE | 1 558 415 | General population (20-80 years) | 1D CNN (ResNet) | MAE: 8.4 ± 7 years (CODE-15% test set) | ECG age gap > 8 years: HR 1.79 for all-cause mortality | Yes (ELSA-Brazil, SaMi-Trop) |
| Sau 2024 | AIRE platform (BIDMC development cohort) | 189 539 | Mixed clinical populations (20-85 years) | 1D CNN + discrete-time survival model | All-cause mortality: C-index 0.64-0.78 | Predicts survival curves, time-to-mortality, future CVD | Yes (5 external cohorts USA, UK, Brazil) |
| Zhang 2023 | UK Biobank | 44 094 | General population (40-75 years) | 1D CNN (ResNet; CODE study) | ECG age modifiable by lifestyle (2.5 years difference) | Favorable lifestyle: 2.5 years younger ECG age | No |

Limitations of ECG clocks

Despite their impressive performance, current ECG-based aging models face several important limitations. While AI ECG models can predict mortality from ECG images with C-indices of 0.77–0.78, performance varies across patient populations and ECG formats [11], highlighting the challenges in generalizability across cohorts and devices. Many AI models are trained on ECGs collected from hospital settings, where patients have a higher prevalence of CVD than population cohorts [10]. When tested on healthier or more diverse populations, the performance of those models drops. Therefore, the models may not perform as well in primary care or community settings with different demographic and disease characteristics. Furthermore, despite progress in interpretability, it remains sometimes unclear which specific ECG features and patterns are the most important for age prediction in these models [15]. Finally, there is a lack of longitudinal validation demonstrating whether improvements in predicted ECG age correlate with reduced cardiovascular events following interventions, making it unclear whether lowering ECG age truly means improving health outcomes [16]. The biological mechanisms underlying accelerated ECG aging remain incompletely understood. While accelerated ECG aging is associated with cardiovascular risk factors [11, 17], the molecular and metabolic drivers of these changes remain

unclear. Most studies use a single-modality approach, relying solely on ECG signals and not integrating complementary biological data. Several molecular clocks have been developed to characterise aging, which could complement or be integrated with ECG clocks.

## Metabolic aging and metabolomic clocks

Because the metabolome lies downstream of the genome, transcriptome, and proteome, it reflects the combined influence of these upstream biological layers. As a result, it provides a dynamic snapshot of an individual's current biological state and is sensitive to factors such as diet, microbiome composition, environmental exposures, and acute physiological changes [18, 19]. Many metabolites are also influenced, or even directly produced, by the gut microbiome. Metabolomic profiles can thus also capture host-microbiome interactions relevant to cardiometabolic health.

Development and performance of metabolomic clocks

Several studies have used metabolomic profiles to build predictive models of chronological age, often referred to as metabolomic clocks. Early work by Hertel *et al.* (2016) demonstrated that serum metabolomics could predict age within about five years, driven by lipid and amino acid metabolism [20]. Robinson *et al.* (2020) extended these findings using untargeted mass spectrometry, identifying systematic age-related shifts in lipids, energy metabolites, and xenobiotics [21]. More recently, metabolomic-based aging scores such as MetaboHealth [22] and MileAge [23] demonstrate strong associations with frailty, all-cause mortality, and chronic disease burden. For example, individuals whose metabolomic age exceeds their chronological age tend to show increased frailty, shorter telomeres (in the case of MileAge), and elevated risk of mortality. Moreover, in comparative assessments, these scores outperform many earlier biological age predictors (especially those trained purely on chronological age) for certain endpoints. An overview of published metabolomic aging clocks, including cohorts, analytical platforms, key metabolites, performance metrics, and validation status, is provided in Table 2.

In larger cohort analyses, such as the Long Life Family Study, hundreds of metabolites were associated with age, longevity, and mortality, particularly those involved in tryptophan metabolism and the tricarboxylic acid cycle [24]. A younger metabolomic age was associated with approximately a 5% lower mortality risk. More recently, a multi-organ model using 107 plasma metabolites from 274,000 UK Biobank participants achieved an MAE of ~ 6 years and revealed strong links between organ-specific metabolomic age gaps and cardiometabolic diseases [25].

Key pathways and microbiome metabolites

Lipid metabolism undergoes substantial changes with aging, with age-associated alterations in lipid profiles contributing to cardiovascular, metabolic, and neurodegenerative disorders [26]. The gut microbiome plays a central role in altering lipid metabolism, thereby influencing age-related changes in circulating lipid metabolites. Comparative studies in mice and humans show that microbiome-derived

metabolites, particularly lipids, increase with age [27]. An example is trimethylamine N-oxide (TMAO), a microbial product of choline and L-carnitine metabolism. Higher TMAO levels are linked to aortic stiffness, hypertension, coronary heart disease, and frailty, acting through oxidative stress and protein crosslinking [28]. Other microbial metabolites, such as short-chain fatty acids, bile acids, and amino acid derivatives, modulate host metabolism and inflammation, and a younger “microbial age” has been associated with reduced cardiovascular risk [29].

**Table 2. Metabolomic aging clocks**.
Overview of major metabolomic aging clock studies and their key characteristics.

| Study | Cohort | Sample Size | Population | Platform/Method | Key Metabolites/ Pathways | Performance | Key Findings | External Validation |
|---|---|---|---|---|---|---|---|---|
| Hertel (2016) | German cohort | ~2 000 | General population (18–80 years) | NMR spectroscopy | Lipids, amino acids | MAE ~5 years | Serum metabolomics predicts age | Internal cross-validation |
| Robinson (2020) | UK Airwave | 2 239 | Working age adults (30–65 years) | Untargeted LC–MS | Lipids, energy metabolites, xenobiotics | Correlation with epigenetic age: r = 0.3–0.4 | Accelerated aging related to obesity, diabetes, alcohol, depression | Yes (Northern Finnish Birth Cohort) |
| Mutz (2024) | UK Biobank | 225 212 | Middle aged/older adults (40–70 years) | 168 plasma metabolites (Nightingale NMR) + ML | 17 key metabolites | MAE 5.31–6.36 years | MileAge strongly associated with frailty, telomere length, chronic illness, mortality | Internal cross-validation, independent cohorts |
| Sebastiani (2024) | LLFS | 52 764 | Older adults & longevity families (24–110 years) | Plasma metabolomics (408 metabolites) | Tryptophan metabolism, TCA cycle | Strong age–metabolite associations ($p < 10^{-45}$) | Younger metabolomic age: ~5% lower mortality risk, longevity pathways | Survival analysis & longevity family design |
| Anagnostakis (2025) | UK Biobank | 274 247 | General population (40–70 years) | 107 plasma metabolites (multi-organ ML model) | Organ-specific metabolites | MAE ~6 years | Multi-organ metabolomic age gaps linked to cardiometabolic disease & higher mortality | Large-scale population validation |

Limitations of metabolomic clocks

Despite these advances, metabolomic clocks face several challenges. Most studies are cross-sectional, meaning they compare different individuals at a single time point rather than tracking the same individuals over time, which limits insight into how biological aging progresses within individuals. Data preprocessing and normalization vary across platforms, reducing reproducibility, while external factors such as diet, medications, and microbiome composition add additional variability. Furthermore, untargeted metabolomics often captures a large proportion of unannotated signals (“dark metabolome”), which may improve predictive performance but limit biological interpretability. In addition, pathway-level interpretation of metabolomic data is limited by incomplete metabolite annotation and inconsistencies in pathway databases, which may affect the robustness of biological pathway enrichment analyses [30]. In contrast, targeted platforms such as NMR-based lipid panels offer higher reproducibility but limited metabolic coverage [31]. Additionally, substantial heterogeneity in metabolite coverage across studies hinders direct comparison and external validation of aging signatures [30]. Despite these challenges, metabolomic clocks provide a promising complement to other biological aging measures and may reveal pathways driving accelerated aging, particularly when integrated with functional and epigenetic data.

# Epigenetic aging and DNA methylation clocks

Epigenetic alterations, particularly DNA methylation changes, are closely linked to aging and provide highly accurate molecular markers of biological age [32]. Over the past decade, epigenetic clocks have demonstrated that DNA methylation patterns can predict chronological age with high precision and link accelerated epigenetic aging to a wide range of age-related diseases and adverse health outcomes [33]. These epigenetic age measures have been consistently associated with morbidity, mortality, and a range of age-related health outcomes [32]. Four major clocks have shaped the field: the Hannum clock (2013, 71 CpG sites in blood, tissue-specific) [34], Horvath clock (2013, 353 CpG sites across 51 tissues, pan-tissue applicability) [35], PhenoAge (2018, integrating clinical biomarkers with methylation to predict morbidity and mortality) [36], and GrimAge (2019, incorporating DNA methylation surrogates for plasma proteins and smoking exposure, the strongest predictors of lifespan and healthspan) [37]. While most first- and second-generation epigenetic clocks were developed using penalized regression approaches such as Elastic Net [33], recent studies have also begun to explore DL and graph-based neural network models to capture nonlinear interactions between CpG sites and potentially improve biological signal extraction [38]. Importantly, while first-generation clocks were primarily optimized to predict chronological age, newer generations of clocks increasingly aim to capture biological aging, morbidity, and mortality risk [33]. The characteristics, training cohorts, performance, and validation of major epigenetic clocks are summarized in Table 3. More recently, third-generation clocks such as DunedinPACE have been developed to quantify the longitudinal pace of biological aging rather than chronological age alone [39].

Clinical relevance and biological mechanisms

Epigenetic age acceleration refers to the deviation between an individual's DNA methylation-predicted age and their chronological age, either calculated as a simple difference or as residuals after regressing DNA methylation age on chronological age. Positive value indicates accelerated biological aging while a negative value indicates decelerated aging. GrimAge shows particularly strong associations with clinical outcomes: each 5-year acceleration corresponds to 20% higher coronary heart disease risk and superior cardiovascular mortality prediction compared to earlier clocks [37]. PhenoAge acceleration similarly associates with heart failure and all-cause mortality independent of traditional risk factors [36]. GrimAge specifically captures methylation patterns associated with inflammatory plasma proteins and smoking, directly linking environmental exposures to biological damage [37]. The methylation sites included in these clocks likely reflect a mixture of processes, including cellular aging, stress responses, and possibly adaptive as well as damaging methylation changes [40].

**Table 3. DNA methylation-based epigenetic aging clocks.**

| Study | Cohort | Sample Size | Population | Clock/Method | Performance | Key Findings | External Validation |
|---|---|---|---|---|---|---|---|
| Hannum (2013) | Whole blood samples (multi-cohort) | 656 | General population (19–101 years) | 71 CpG sites: Elastic Net | MAE ~4–5 years | Accurate blood-specific age prediction | None in the original study, subsequently validated in independent cohorts |
| Horvath (2013) | Multi-tissue datasets | 8 000 | Diverse populations (0–101 years) | 353 CpG pan-tissue DNAm clock | MAE ~3.6 years | Universal epigenetic aging marker across tissues | Yes<br>Multi-cohort, multi-tissue |
| Levine (2018) (PhenoAge) | NHANES III | 9 926 | General population (20–90 years) | Elastic Net + clinical biomarkers | MAE ~4.5–5 years | Predicts morbidity, CVD, and all-cause mortality | Yes<br>Multiple independent population cohorts (NHANES, WHI, FHS) |
| Lu (2019) (GrimAge) | Framingham Heart Study Offspring | 2 356 | Middle-aged and older adults (40–92 years) | DNAm surrogates for plasma proteins + smoking | Strongest mortality prediction (HR ≈ 1.08–1.10 per year acceleration) | Best predictor of lifespan, healthspan, and CVD mortality | Yes<br>Multiple large independent cohorts with mortality outcomes (FHS, WHI, JHS) |

Comparison of major DNA methylation-based epigenetic aging clocks.

Importantly, different epigenetic clocks are only moderately correlated with one another, and epigenetic age acceleration measures derived from different clocks often show substantial variability. This suggests that individual clocks capture distinct aspects of the aging process rather than a single universal biological age [39]. First-generation clocks such as Horvath and Hannum were trained primarily to predict chronological age, whereas second-generation clocks such as PhenoAge and GrimAge were optimized to capture physiological decline, disease risk, and mortality [41]. As a result, these clocks may reflect overlapping but biologically distinct aging pathways [42]. Furthermore, correlations between epigenetic clocks and aging measures derived from other omics layers, including metabolomics and proteomics, are generally modest, supporting the concept that biological aging is a multidimensional process that cannot be fully captured by a single biomarker [43].

Limitations of epigenetic clocks and need for multi-omics integration

Despite their predictive power, epigenetic clocks face important limitations. Most clocks were developed using DNA methylation data derived from blood, yet aging trajectories may differ substantially across tissue, including cardiac tissue. Population generalizability remains unclear, as training cohorts predominantly comprised European-ancestry participants. Technical factors, including batch effects and normalization methods, further complicate cross-study comparisons. Most critically, it remains unclear whether age-associated DNA methylation changes play a causal role in aging or primarily act as biomarkers of cumulative exposure and damage. This distinction is important as causal mechanisms may represent therapeutic targets, whereas non-causal biomarkers are mainly useful for risk stratification and disease monitoring.

Beyond these limitations, several critical questions remain unanswered in cardiac aging research. For example, it is unclear whether individuals with accelerated epigenetic aging also exhibit accelerated ECG aging, or whether these measures reflect distinct biological processes acting at different levels of cardiac aging. Similarly, the relationship between epigenetic aging and metabolomic aging remains incompletely characterized. While modest correlations between metabolomic and epigenetic age

estimates have been reported (r = ~ 0.3–0.4), these findings suggest that each clock captures partly independent aspects of biological aging [21]. Certain microbial-derived metabolites, such as TMAO, are of particular interest because they link gut microbiome activity, systemic metabolism, and cardiovascular pathology. While initial studies have begun to explore such relationships, comprehensive integration of molecular, metabolic, and functional cardiac aging measures remains limited. These gaps highlight the need for longitudinal multi-omics studies to resolve the tissue specific and molecular networks underlying cardiovascular aging.

## Integrating Multi-Omics and functional cardiac data with AI models

While the three types of clocks described above partially overlap, reflecting shared underpinning aging mechanisms, each captures a distinct aspect of biological aging. Epigenetic clocks quantify molecular and cellular aging that accumulate over time in given tissues and reflect lifelong environmental and physiological exposures [36]. Metabolomic clocks characterize physiological status at a given time and systemic aging by capturing modifications in metabolic pathways, oxidative stress, inflammation, and gut microbiome activity [44]. ECG-based clocks assess organ-level functional aging by tracking how the heart deteriorates with age [45]. Together, these clocks provide complementary perspectives on aging. Their integration would provide multiscale insights into aging biology (molecular → systemic → functional), capture interactions between metabolic and cardiovascular aging, and suggest explanations for why some individuals age faster than others. Emerging evidence also suggests biological links between epigenetic and metabolic aging processes [40].

Data integration and standardization

Multi-modal data integration requires harmonizing fundamentally different data types. ECGs capture millisecond-scale electrical activity, while metabolomic datasets reflect slower chemical and physiological changes over days to months, and epigenetic clocks reflect changes accumulated over a lifetime. Each platform, e.g., UHPLC-HMRS for metabolomics, methylation arrays for epigenetics, and various ECG recording devices, introduces unique sources of noise and batch effects. Standardized protocols, reference materials, and shared quality control pipelines are needed to ensure consistency across data modalities. Missing data and timing mismatches between measurements (e.g., ECG and blood sampling on different days or at different times of day) further complicate integration, underscoring the need for careful study design and robust imputation strategies.

Challenges and opportunities of multimodal integration

Integrating ECG-based, metabolomic, and epigenetic aging clocks offers an innovative framework for precision medicine, although several methodological, biological, and translational barriers must be overcome before reaching the clinics.

Methodological challenges

The high dimensionality and heterogeneous structure of ECG, metabolomic, and methylation data complicate model development. Computational approaches such as DL can learn shared representations across modalities, but these methods require large, well-annotated datasets and are susceptible to overfitting when sample sizes are limited. Different fusion strategies exist [46], including feature-level fusion (combining all variables into a single model input), intermediate fusion (learning shared representations across modalities), and decision-level fusion (combining predictions from separate models). Each approach involves trade-offs between predictive performance, interpretability, and robustness to missing data. Moreover, multimodal models may reduce transparency, making it harder to attribute predictions to biologically meaningful features.

### Opportunities

Despite these challenges, integration offers important advantages. Combining molecular, metabolic, and functional information may improve risk stratification and provide a more comprehensive characterization of biological aging. Discordance between clocks could identify individuals whose specific organs are aging faster than other systems, or whose overall physiology compensates for these local changes. Integrated models may also generate mechanistic hypotheses by linking molecular signatures to functional cardiac phenotypes, thereby bridging molecular biology and clinical cardiology. Future studies should explore how ECG-based models can be integrated with metabolomic and epigenetic data in longitudinal cohorts to determine whether multimodal aging profiles improve the prediction of cardiovascular events beyond single-modality approaches.

### Generalizability across populations and technologies

The performance of aging clocks often varies across populations and devices. AI-ECG models that predict mortality with C-indices of 0.77–0.78 [11] may show less reduced performance on different ECG devices, lead configurations, or populations with distinct disease prevalence and ancestry. Diet, lifestyle, and geography alter metabolomic and microbiome-based profiles, impacting model outputs and weakening transferability [11]. Similarly, epigenetic clocks can show reduced accuracy in populations underrepresented in training cohorts or when used on tissues other than those used for model development. These challenges highlight the need for population- and device-agnostic models, standardized preprocessing pipelines, open data sharing, and external validation across diverse cohorts to enable reliable performance and clinical adoption.

### Interpretability and mechanistic understanding

Despite progress in explainable AI, the physiological relevance of aging clock predictions remains only partially understood, as current methods provide limited insight into both the ECG features driving model outputs and their underlying biological meaning. Weak correlations among age-associated metabolites indicate that metabolomic clocks capture age through complex, partially independent, metabolic pathways rather than single biomarkers [20]. When it comes to epigenetic clocks, the CpG sites used to estimate biological age differ across tissues and are not necessarily located within genes directly

involved in aging processes. Instead, many of these CpGs reflect broader patterns of chromatin accessibility and gene regulatory activity that change with age in a tissue-specific manner [35]. While explainability tools showed that ECG clock models attend to known age-related ECG-features, how these relate mechanistically to metabolic or epigenetic aging remains uncertain. Longitudinal studies are needed to clarify whether discordance between ECG age, metabolomic age, and epigenetic age reflects organ-specific aging or compensatory metabolic reserve.

## Longitudinal and interventional evidence

A critical challenge facing aging clock research is the predominance of cross-sectional study designs, in which biological age is inferred from single time-point measurements rather than repeated assessments within the same individuals. As a result, most existing aging clocks cannot determine whether changes in biological age reflect true alterations in aging trajectories or transient, reversible biomarker fluctuations. This limitation makes it difficult to assess whether aging clocks predict future age-related health outcomes or merely correlate with current health status [43]. While pilot randomized controlled trials have demonstrated preliminary evidence that diet and lifestyle interventions can reduce epigenetic age (for example, an 8-week intervention resulted in a 3.23 year decrease in epigenetic age compared to controls [47]), linking changes in DNA methylation-based age estimates or CpG methylation patterns to clinically meaningful long-term health outcomes remains challenging. To establish the translational value of aging clocks, randomized controlled trials are needed to simultaneously assess whether interventions that reduce biological age also prevent age-related diseases and mortality. Additionally, distinguishing whether molecular biomarkers represent true causal drivers of aging or merely correlate with age remains essential for therapeutic translation, as data-driven algorithms often detect a mixture of features that can cause aging, features caused by aging, and uninformative correlates of chronological age.

## Clinical Translation and Equity

For aging clocks to inform personalized medicine, models must not only predict risk but also provide actionable insights. Clinicians need evidence-based guidance on how to respond when biological age exceeds chronological age through intensified screening, lifestyle interventions, or targeted therapy. Integrating multi-omics and functional aging metrics into clinical workflows will require regulatory validation, cost-effectiveness analyses, and interoperability with electronic health record systems. Finally, ensuring equitable access is important. Metabolomic and epigenetic profiling remains costly and technically demanding. Beyond assay cost, clinical deployment also requires trained personnel, computational infrastructure, and standardized interpretive frameworks, as model outputs must be translated into clinical decisions by physicians whose training and experience may influence downstream care. This raises concerns about accessibility in low-resource settings. Affordable assays, open-source computational tools, and validation across diverse populations will be essential to ensure that precision aging medicine benefits patients globally.

## Conclusions and perspectives

The convergence of AI with ECG, metabolomic, and epigenetic data provides a powerful framework to capture biological aging across molecular, systemic, and functional levels. These complementary aging clocks offer distinct but overlapping insights into how individuals age, revealing potential mechanisms that drive organ-specific vulnerability or resilience. Understanding concordance and discordance between different aging measures may uncover unique aging phenotypes and identify modifiable biological pathways. Moving forward, integrating these data layers within longitudinal, population-diverse cohorts will be essential to develop interpretable, generalizable models that can guide proactive, personalized interventions. Ultimately, such integration approaches have the potential to shift precision medicine from reactive disease management to strategies aimed at maintaining healthspan and preventing cardiovascular disease, enabling more effective, mechanism-informed interventions for healthy aging.

## Abbreviations

AI
Artificial intelligence
AIRE
AI-ECG Risk Estimation
CNN
Convolutional neural network
CVD
Cardiovascular disease
CpG
Cytosine-phosphate-guanine dinucleotide
ECG
Electrocardiogram
HR
Hazard ratio
DL
deep learning
UHPLC-HMRS
Ultra High-Performance Liquid Chromatography-High-Resolution Mass Spectrometry
LVEF
Left ventricular ejection fraction
MAE
Mean absolute error
ML
Machine learning
ResNet

Residual neural network
TMAO
Trimethylamine N-oxide

# Declarations

## Availability of data and materials

Data sharing is not applicable to this article as no datasets were generated or analysed during the current study.

## Competing interests

The authors declare that they have no competing interests.

## Funding

This work was supported by a CNRS doctoral contract within the International Research Project (IRP) Metabo-LIC directed by Marc-Emmanuel Dumas, in collaboration with the U1283/UMR8199 laboratory at the University of Lille and Imperial College London. NG benefits from a STaRS funding from the Conseil Régional des Hauts-de-France (24005486). DPO'R is funded by the Medical Research Council (MC_UP_1605/13); National Institute for Health Research (NIHR) Imperial College Biomedical Research Centre; and the British Heart Foundation (RG/F/24/110138, RE/24/130023, CH/F/24/90015). AK is supported by the EFSD / Novo Nordisk Foundation Future Leaders Award (Grant number: NNF25SA0106140).

## Author's contributions

SÅ conducted the literature review, drafted the manuscript, and created the figures and tables. MED and NG supervised the work and critically revised the manuscript. All co-authors provided intellectual input, reviewed the manuscript, contributed comments and revisions, and approved the final manuscript.

## Acknowledgements

The authors thank the members of the U1283/UMR8199 laboratory at the University of Lille and the CNRS-ULille-Imperial Metabol-IC team for helpful discussions and support.

## Ethics approval and consent to participate

Not applicable (review article).

## Consent for publication

Not applicable (review article).

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# Figures

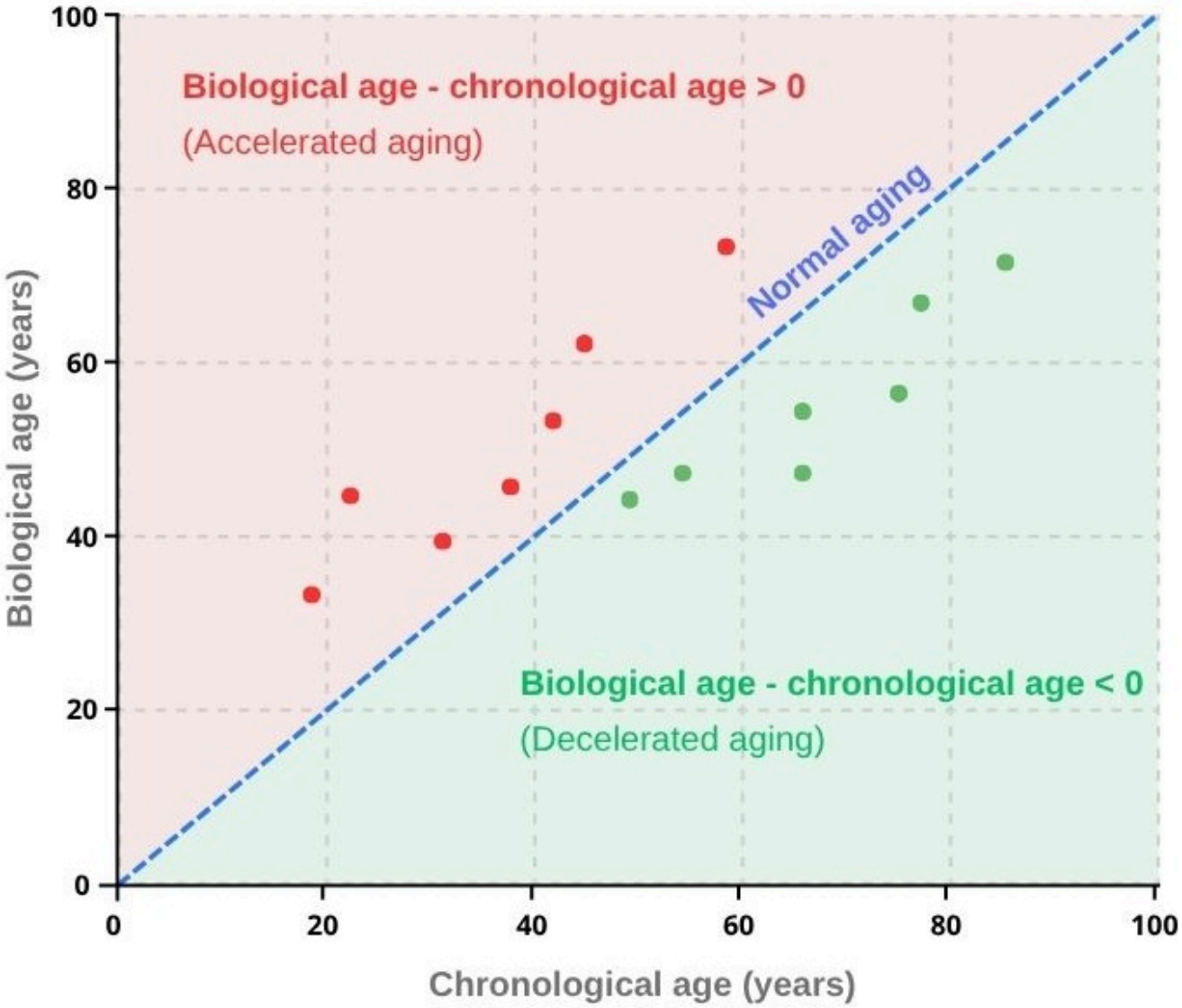
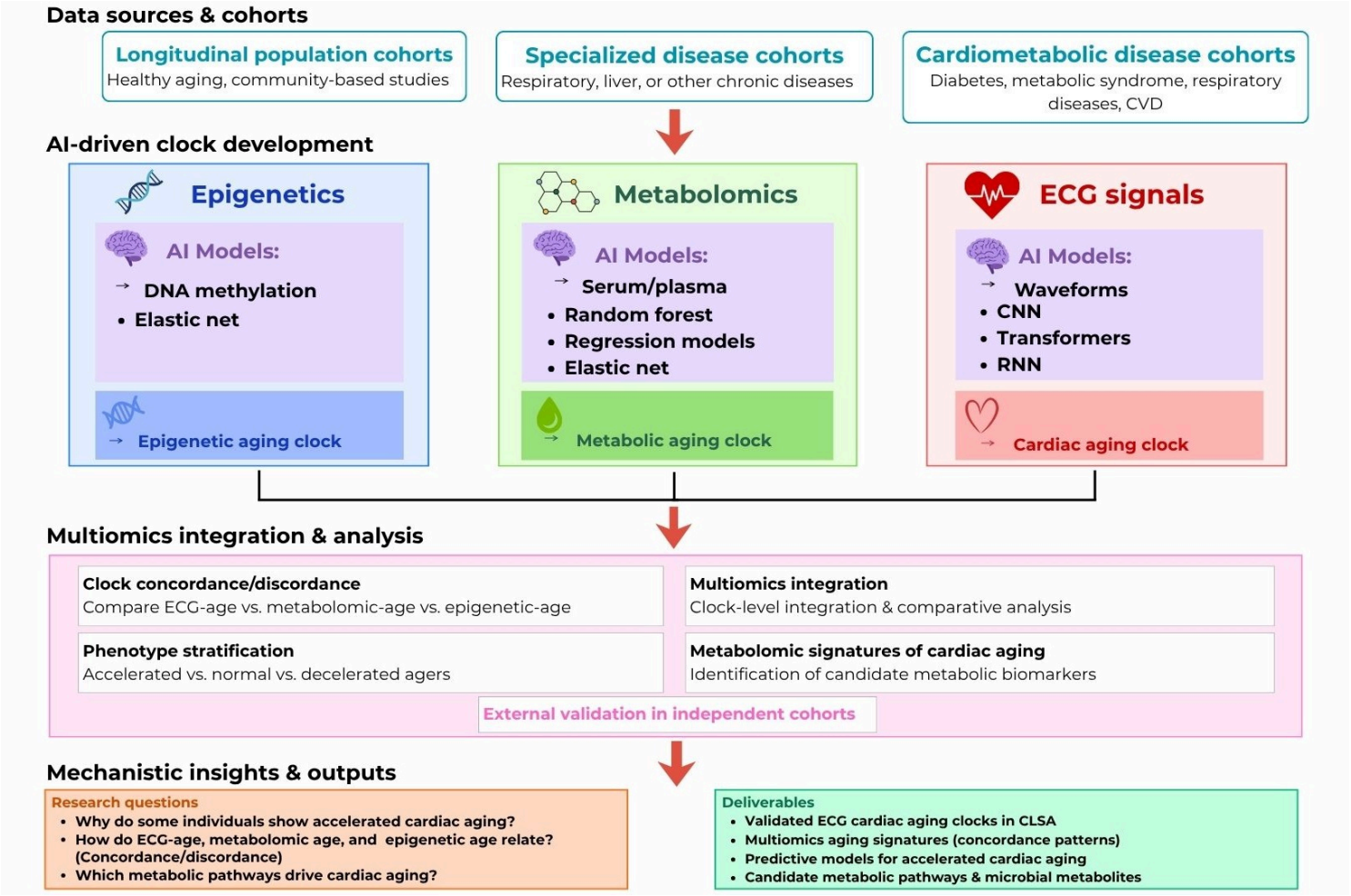



**Figure 1**

**Relationship between chronological age and biological age.**

Each point represents an individual. The dashed line indicates where biological age equals chronological age; points above the line correspond to accelerated aging, and points below indicate decelerated aging.




## Figure 2

### Conceptual framework for multi-omics integration in cardiac aging research

Schematic overview of how complementary biological layers can be used to estimate cardiac aging. Functional cardiac aging may be assessed using ECG signals, while systemic metabolic aging and molecular/cellular aging can be captured through metabolomic and epigenomic data, respectively. Aging clocks can be developed independently within each modality using AI or ML approaches and subsequently integrated through statistical or computational frameworks to identify shared aging patterns, candidate biomarkers, and links to cardiovascular risk.