Review

Integrated Multi-Omics for Novel Aging Biomarkers and Antiaging Targets

Lei Wu [1,2,†], Xinqiang Xie [1,†], Tingting Liang [1,2,†], Jun Ma [2], Lingshuang Yang [1], Juan Yang [1,2], Longyan Li [1], Yu Xi [1], Haixin Li [1], Jumei Zhang [1], Xuefeng Chen [2], Yu Ding [3,*] and Qingping Wu [1,*]

1 Guangdong Provincial Key Laboratory of Microbial Safety and Health, State Key Laboratory of Applied Microbiology Southern China, Institute of Microbiology, Guangdong Academy of Sciences, Guangzhou 510070, China; wuleigdim@163.com (L.W.); woshixinqiang@126.com (X.X.); gdim_liangtt@outlook.com (T.L.); yangls8272@163.com (L.Y.); yj18185238563@163.com (J.Y.); 18868006204@163.com (L.L.); xiyu_0604@163.com (Y.X.); 201920146453@mail.scut.edu.cn (H.L.); zhangjm926@126.com (J.Z.)
2 School of Food and Biological Engineering, Shaanxi University of Science and Technology, Xi'an 710021, China; nwu_majun@hotmail.com (J.M.); chenxf@sust.edu.cn (X.C.)
3 Department of Food Science and Technology, Institute of Food Safety and Nutrition, Jinan University, Guangzhou 510632, China
* Correspondence: dingyu@jnu.edu.cn (Y.D.); wuqp203@163.com (Q.W.); Tel.: +86-020-87688132 (Q.W.)
† These authors contributed equally to this work.

Abstract: Aging is closely related to the occurrence of human diseases; however, its exact biological mechanism is unclear. Advancements in high-throughput technology provide new opportunities for omics research to understand the pathological process of various complex human diseases. However, single-omics technologies only provide limited insights into the biological mechanisms of diseases. DNA, RNA, protein, metabolites, and microorganisms usually play complementary roles and perform certain biological functions together. In this review, we summarize multi-omics methods based on the most relevant biomarkers in single-omics to better understand molecular functions and disease causes. The integration of multi-omics technologies can systematically reveal the interactions among aging molecules from a multidimensional perspective. Our review provides new insights regarding the discovery of aging biomarkers, mechanism of aging, and identification of novel antiaging targets. Overall, data from genomics, transcriptomics, proteomics, metabolomics, integromics, microbiomics, and systems biology contribute to the identification of new candidate biomarkers for aging and novel targets for antiaging interventions.

Keywords: aging; aging biomarkers; antiaging targets; multi-omics; aging clock

1. Introduction

In 2019, there were an estimated 702 million people aged ≥65 years according to world population prospects 2019: Highlights, accounting for 9.1% of the world population. The aged population also grows at approximately 3% per year. In addition, human life expectancy rapidly increases, i.e., from 64.2 years in 1990 to 72.6 years in 2019, and is predicted to increase further to 77.1 years in 2050 [1]. Thus, the risk of developing aging-related diseases increases.

Aging is a physiological process in organisms in which multifactorial processes, including genetic factors, external environmental stimuli, and lifestyle factors, determine a progressive decline over time. Environmental factors may have a cumulative and multiple impact on health and longevity. The idea of "healthy lifestyles and environments" comes from the observation of geographical clusters of centenarians around the world, with five identified "longevity hotspots" known as Blue Zones, which are located in Sardinia (Italy), Okinawa (Japan), Loma Linda (California), Nicoya (Costa Rica), and Ikaria (Greece). Thus, their lifestyles and environments are possibly more conducive to longevity than those of

the rest of the world. The populations in these areas are characterized by having an active, stress-free lifestyle, strong community bonds, and spirituality. Maybe these are exactly what we in the "non-blue zone" want to learn. It is also subject to regional restrictions such as lifestyle, economic conditions, and geography; they cannot necessarily be broadly extrapolated. However, among tissues and organs, different individuals age at different rates. The aging rate is highly variant, and these specific changes often affect organ functions [2,3]. The aging of the physiological systems and the changes in their functions lead to various chronic diseases and metabolism-related syndromes [4,5].

Therefore, the characterization of aging-related biomarkers is expected to pave the way for the discovery of novel antiaging targets [6]. Understanding the causes of aging and disease and the relationship between the two is important for aging biomarkers that promote the development of geriatrics and clinical translation. To date, there is no accurate independent aging biomarker that can accurately reflect the aging state or aging rate of people. Aging can be characterized by biomarkers [7]. To achieve this goal, studies at the multi-omics level, which integrates epigenomics, transcriptomics, proteomics, metabolomics, and microbiomics data, can provide a more comprehensive overview [8].

Due to the rapid development of bacterial species resource banks and large biological databases around the world, the use of bioinformatics has remarkably improved, and multi-omics methods are the most promising [9]. Currently, various national projects for the biobanking of samples obtained from many people for subsequent omics analysis exist. For instance, biobanks of aging research omics projects include MARK-AGE [10], EUROBATS, and UKBiobank [11]. Similarly, artificial intelligence will greatly deepen our understanding of aging in the near future and transform the most reliable method of assessing biological age into clinical practice. Previously, we have attempted to characterize the gut microbiota of centenarians to determine whether a relationship between the gut microbiota and human longevity exists. Using 16S rRNA gene and metagenomic sequencing methods, we constructed the longest human gut microbiota trajectory of aging and analyzed the composition and function of the gut microbiota in 247 healthy subjects aged 0–110 years. We also conducted in vitro and in vivo experiments in mice.

The multi-omics approach has become the gold standard in different fields of biological sciences. The multi-omics approach can increase the number of identified markers for aging biomarkers with novel insights into aging and of novel targets for antiaging. In this review, we summarize recent studies in the field of aging based on different multi-omics viewpoints, i.e., genomics, transcriptomics, proteomics, metabolomics, and microbiomics. We integrate aging biomarkers of different omics levels to better discover novel targets for antiaging interventions. These promising aging biomarkers could be useful for clinical research.

2. The Necessity of Distinguishing Chronological Age and Biological Age

Without a method to assess the personal aging rate, determining preventive interventions for aging is impossible. Aging biomarkers show the changes in the molecules, cells, and organs of the human body with age. Ideally, these biomarkers should slow their progression with age or reverse to a young state [12]. Chronological age represents a person's actual age and is calculated based on the time elapsed in a person's life [13]. Biological age refers to an individual's overall health status at a certain point in time of physiological age. Generally, the environment, diet, life, and psychological factors should be considered. Biological age has been revealed as a better predictor than chronological age, and its measurement can facilitate the assessment of colonoscopy-related colorectal adenoma risk [14,15].

In aging research, it is common to use chronological age. However, due to the heterogeneity of aging, chronological aging is unpractical. There has been a discrepancy between the predictions of biological age and chronological age. Considering the heterochronism of aging, the measurement of biological age becomes complicated, as it involves the calculation of many target molecules that indicate the dynamics of different processes. The panels of biomarkers can act as an integrated tool for measurements. Generally, special indicators

are used to accurately indicate biological age. Klemera and Doubal used chronological age as one of the biomarkers, which is the most popular biomarker [16,17]. The principal component analysis method unites equation construction, correlation analysis, and redundancy analysis [18,19]. The main problem with biological age assessment is that the function of chronological age is unknown among the different available measurement methods. Some people believe that it is a very important biomarker [20], while others consider that the aging rate does not need to be measured by chronological age [21].

Advances in artificial intelligence and statistics provide opportunities to accurately estimate biological age. However, they are not fully effective against heterogeneous populations, and there is no clinical certainty. The application of some aging biomarkers from different sources leads to a reduction in the resolution of most biomarkers. Since the ideal biological age estimation method should be comprehensive and complete, we suggest an integrative approach based on multi-omics technologies for aging biomarkers and novel antiaging targets. This multi-omics method is based on the multi-layered organizational logic of life, thus making the prediction of biological age more accurate.

3. Multi-Omics for Aging Clocks

3.1. Epigenetics Aging Clocks

Biological age estimation based on DNA methylation has been accurately discussed [22–24]. As an "age estimator", the epigenetic aging clock is used to estimate the epigenetic (biological) age of DNA. It also demonstrates that age-related diseases are associated with higher biological age relative to the chronological age. This phenomenon is called epigenetic age acceleration [22].

The discovery of some aging clocks can predict age-related pathologies, such as cancers, heart disease, and diabetes [25]. There are other types of epigenetic clocks from whole blood [26–30], skin [31], and saliva [32]. The aging clock known as DNA methylation GrimAge is an instrument that allows us to view epigenetic acceleration of aging from a new perspective. It can predict the time-to-death and comorbidity count, time-to-cancer, and time-to-coronary heart disease [33]. In a biological aging clock based on DNA methylation, the main indicator of biological age is the methylation of ribosomal DNA exclusively. It can accurately characterize the biological age and show the organism's response to the treatment of aging and effective antiaging interventions [34].

3.2. Transcriptomics Aging Clocks

The positive increase in age reflects faster biological aging. Peters et al. conducted a meta-analysis of 7074 individual peripheral blood samples, in which 11,908 genes were characterized to create age-related predictors. They found that the average absolute error between the chronological age and the predicted age was 7.8 years [35]. Another important study focused on the aging of the transcriptome of skin cells served as a pioneering method for determining the biological age of such datasets [36]. The age was predicted by linear discriminant, and the median absolute error and average absolute error of 4 years and 7.7 years were obtained, respectively [36,37].

The aging rate varies greatly between individuals and groups and will be significantly affected by factors such as genetics, environment, lifestyle, etc. Due to the data type, transcriptome aging clocks have weaker correlations with the chronological age than DNA methylation aging clocks [24,38]. To address this, a standardized cohort is needed. In a study of 6465 individual blood samples collected from 17 datasets, the differences in technical performance had a more significant effect on blood expression profiles than disease and age itself [39]. Then, cross-platform normalization methods, normalization through reference genes, distribution transformation, and quantile normalization were used to successfully eliminate the batch processing effects. A deep neural network was utilized as a predictive index to yield an average absolute error of 6.14 years and a Pearson correlation accuracy of 0.91 [39].

In summary, the biological age prediction technology based on transcriptomics has developed rapidly, and its accuracy level has been continuously improved. Thus, transcriptomics aging clocks will catch up with methylation aging clocks in the near future.

3.3. Proteomics Aging Clocks

To help optimize aging clocks and determine the potential novel targets for antiaging interventions, proteomics aging clocks have been systematically reviewed and analyzed. Proteins are studied, because they significantly change their expression levels with age and represent functional products, unlike transcriptome changes, which are not always associated with proteome changes [40,41]. Since previous proteomics studies have used various proteomics techniques, sample sizes and types, and statistical methods, significant differences in the results have been observed. Even when analyzing the same biological sample, the findings can be quite different [42,43].

To achieve these goals, a systematic review of 36 different proteomic analyses was performed, each of which identified proteins that changed significantly with age [44]. There were 32 proteins that have been reported at least five times and 1128 at least twice. Each of these 32 proteins is related to aging and age-related diseases. Furthermore, 1128 common proteins associated with gene regulation, extracellular matrix, and inflammation were analyzed based on bioinformatics enrichment. Finally, a new proteomics aging clock was proposed, which is composed of three or more proteins in the plasma that change with age in different studies. Using a large patient cohort of 3301, the proposed proteomics aging clock was confirmed to accurately predict the age of a person [44].

Another study analyzed 2925 plasma proteins in a cohort of 4263 subjects and developed a new bioinformatics method. This study revealed significant nonlinear changes in the human plasma proteome with age. Changes in the proteome reflect the different biological pathways and reveal the various genome and proteome associations with age-related diseases and phenotypic traits. This new method of studying aging may provide potential novel targets for age-related diseases [45].

3.4. Metabolomics Aging Clocks

Hertel et al. [46] proposed the use of metabolomics for biological age prediction, called the "metabolomic aging clock". They based their analysis on urine data obtained through ^{1}H-NMR spectroscopy. The metabolomics aging clock can predict the prognosis of weight loss in bariatric surgery patients and can be applied to other fields of medicine. Similarly, van den Akker et al. [47] developed an innovative, biological age measurement method based on metabolomics and analyzed the ^{1}H-NMR serum metabolomics data. To estimate chronological age, they used a linear model trained with metabolomic variables. Finally, they constructed a score reflective of an individual's biological age called metaboAge and showed that the excess of metaboAge over chronological age corresponded with a poor cardiometabolic health.

3.5. Microbiomics Aging Clocks

Using the microbiome aging clock to predict biological age is a relatively new analytical method. However, this method has two problems: one is to find people with similar lifestyles, and the other is to normalize the dataset. Under normal circumstances, the structure and composition of the human gut microbiota will decrease with age; however, the elderly occasionally exhibit a microbiota structure similar to that of adults. The gut microbiome is mainly composed of four phyla: Firmicutes, Proteobacteria, Bacteroidetes, and Actinobacteria [48]. During aging, the relative abundances of *Bifidobacterium*, *Bacteroides*, *Lactobacillus*, *Ruminococcus*, and *Bacillus* decrease, whereas those of *Streptococcus*, *Enterobacter*, *Clostridium*, and *Escherichia* increase [49,50]. The results of studies on aging-related microbial communities are similar to those in microbial communities. In addition to transcriptomics studies, microbiology studies also heavily rely on methodology [51].

In the transcriptional microbiology of aging, the concentration of short-chain fatty acid products in the gut of aging people is low, and it is related to the increase in the number of pathogenic and gas-tolerant bacteria, whose reproduction can lead to malnutrition and age-related diseases [49]. Based on the metagenomic dataset and using deep neural network methods to determine the biological age, including 1673 microbial taxa. An average absolute error of 3.94 years, which is remarkably close to Horvath's [38] 3.4 years, and an R^2 value of 0.81 were obtained. This is the first study to establish a quantitative model of gut microflora aging [49].

4. Multi-Omics Approach for the Discovery of Aging Biomarkers

Aging is the main risk factor for chronic diseases that limits a healthy lifespan. Therefore, the mechanism of aging is a potential therapeutic target. Age correlation analyses involve large amounts of data obtained from various omics analyses, such as genomics (epigenomics), transcriptomics, proteomics, metabolomics, and microbiomics. The main advantages of this method include the analysis of all possible data pertaining to a single person or a large group of people, as well as the common and individual characteristics from a multi-dimensional perspective and the identification of aging markers and novel antiaging targets. Machine learning methods based on deep neural networks are the latest and most complex methods for identifying human aging biomarkers. They can utilize any type of omics data to predict age.

4.1. Aging Genomics

4.1.1. Aging Epigenomics

Epigenetics is the study of changes in the biological phenotype without change alterations in the intrinsic genotype [52], and these changes are mainly caused by the environment [53]. The DNAm model is the most studied epigenetic feature [54–56]. The epigenetic aging clock is a useful predictor of age-related diseases. Most studies on DNAm patterns analyzed peripheral blood samples and showed that the over- and undermethylation of CpG sites are related to mortality. A total of 353 CpG sites can be used to estimate physiological aging [56]. On the other hand, the immune system status can be characterized by 73 CpG sites [23,24], and 10 CpG sites can be used as predictors of cancer and cardiovascular disease mortality [28]. DNAmGrimAg has been correlated with diseases and can predict mortality [33].

The indicators of epigenetic aging are also related to neurodegenerative diseases. For example, Parkinson's disease (PD) is associated with the first acceleration of epigenetic aging clocks [57]. Higher epigenetic age (increased DNAmAge) corresponds with a higher risk for cancer and age-related cartilage degenerative diseases [58,59]. Exercise can improve the DNAm of sarcopenia-related genes, in which the epigenetic aging clock is lower [60]. In addition, the epigenetic aging clock increases with BMIs in patients with obesity and metabolic syndrome [61], indicating the relationship between the epigenetic clock and lifestyles.

4.1.2. Aging Gene Expression

Further, overexpression of the Forkhead box O3 gene (*FOXO3*) in model organisms is related to a prolonged lifespan. *FOXO3* overexpression in the adipose tissue of *Drosophila* [62] and mice results in an extended lifespan [63]. Polymorphisms in the *FOXO3* gene in humans are also associated with longevity [64]. Furthermore, the apolipoprotein E gene (*APOE*) encodes a major cholesterol carrier that supports regulation of the cholesterol and lipid metabolism and cell repair [65]. Furthermore, knocking out the tumor suppressor gene *P53* in mice results in premature aging, organ atrophy, osteoporosis, and a poor antistress response [66].

4.1.3. Telomere-Based Biomarkers

Telomeres are the protective caps of nuclear proteins at the end of eukaryotic chromosomes, composed of repeated TTAGGG. They have the functions of controlling cell and human aging and protecting chromosomes [67]. Telomere length is a typical indicator of biological aging, and telomerase is the main regulator of telomeres [68]. Cell division is accompanied by the shortening of telomere and gradually leads to chromosomal instability with age [69]. In one study involving the systemic knockout of mouse telomerase subunits, a decrease in telomere length, an acceleration of organ dysfunction, and a shortened lifespan were observed [70,71]. The reintroduction of telomerase has great potential for reversing aging [72].

Telomere wear may increase the risk of aging-related diseases [73]. A study on a large population (*n* = 105,539) showed that women have longer telomeres than men and that there are gender-related differences in biological aging [74], which may be due to hormonal differences, such as estrogen levels and the role of the X chromosome [75,76]. There is also a relationship between shorter telomeres and higher mortality [77,78]. Decreased age-related immune surveillance and increased inflammation are associated with the shortening of telomeres and decreased telomerase activity [79]. The telomere length of white blood cells in patients with heart failure was reduced by nearly 40% [80] and, in AD patients, was often shorter [81,82]. Telomere shortening is related to diseases caused by oxidative stress, including Alzheimer's disease [83], diabetes, and cardiovascular diseases, as well as the proinflammatory cytokine tumor necrosis factor alpha [84,85]. An important factor of aging may be the accumulation of reactive oxygen species (ROS) [86]. Notably, the oxidative stress associated with mitochondria also plays an important role. Particularly, carbonyl cyanide-4 (trifluoromethoxy) phenylhydrazone, the uncoupling agent of oxidative phosphorylation in the mitochondria, can depolarize the mitochondria. Mitochondrial dysfunction can increase ROS production, telomere wear, and genome instability [87]. Moreover, cancer cells can proliferate indefinitely due to their ability to maintain a telomere length [88]. However, the relationship between telomere length and cancer susceptibility remains unclear, because cancer types have distinct characteristics [89]. Additionally, it should be further studied whether telomeres and telomerase can be used as direct biomarkers of aging diseases. Specific drugs or nutraceutical methods that can maintain the great conditions of telomere and telomerase without causing adverse reactions should also be explored [90]. In Table 1, we summarize the main potential aging biomarkers identified in genomics studies.

Table 1. Potential aging biomarkers identified in genomics studies.

Omics	Biomarkers	Function/Application	References
Genomics	DNA methylation aging clocks	Biological age estimation method	[38]
	DNA methylation GrimAge	Been correlation with diseases and can predict mortality	[57]
	DNAm pattern of 353 CpG sites	Estimate physiological aging	[56]
	73 CpG sites	Immune system	[23,24]
	10 CpG sites	Predictor of cancer mortality and cardiovascular disease	[28]
	The increase in DNAmAge	Cancer, age-related cartilage degenerative diseases, and tumor tissues	[58,59]
	Forkhead box O3 gene (*FOXO3*)	Related to prolonged lifespan	[62–64]
	The apolipoprotein E gene (*APOE*)	Regulation of the cholesterol and lipid metabolism and cell repair	[65]

4.2. Aging Transcriptomics

4.2.1. Transcriptomics-Based Biomarkers

Deep changes in the transcription profile occur in human aging processes. Another typical example of agingomics is transcriptomics, which studies mRNA groups, including lncRNAome, circRNAome, and exosomal RNAome. Due to the diversity of the methods and goals in each field, there is no unified way to achieve a clear overall view. Visualizing the complete transcriptome also remains a challenge. Focus on research may affect transcriptomics and assist doctors in selecting appropriate biomarkers from various RNA types [91–94]. At present, the characteristics of six gene expression markers of cell senescence have been identified by Frenk & Houseley [95].

As a regulator of lipid homeostasis, phospholipid transport, and macrophage activity, ABCG1 mediates the pathway of endothelial cholesterol efflux and protects blood vessels from chronic inflammation. Such alleles usually determine the human lifespan [96–98]. A study of the human whole-blood transcriptome including 1016 people aged 70–80 years showed that *BIRC2* is an apoptosis regulator of inflammation, cell proliferation, and mitotic kinase signal transduction and was the most downregulated during aging [99]. In another study analyzing whole-blood samples, aging was positively correlated with the expression of 11 genes, namely *AMZ1*, *MANEAL*, *PARP3*, *KIAA0408*, *ISM1*, *CRIP1*, *NEFL*, *PHLDA3*, *DDB2*, *CHN1*, and *CAPN2*, whereas it was negatively correlated with that of four genes, namely *MXRA8*, *SLC4A10*, *CD248*, and *PLEKHA7* [100].

Five transcriptional biomarkers that can distinguish between <65 years old and ≥75 years old have been accurately determined, thereby demonstrating that transcriptomics can classify the elderly [101,102]. The expression of age-related genes can be used to identify aging biomarkers.

4.2.2. MiRNAs, lncRNAs, and circRNAs-Based Biomarkers

MiRNAs are 21–25 nucleotides in length that are involved in biological regulation processes [103,104]. To identify transcriptome-specific biomarkers, the correlation between microRNA expression profiles and chronological age is analyzed. For instance, the expression of miR-22-3p and miR-28-3p are positively correlated with age, whereas that of miR-425-3p, miR-182-5p, and miR-99b-5p are negatively correlated [92]. MiRNA is related to many diseases, such as cancer [105,106], cardiovascular diseases [107,108], hypertension [109], obesity [110], and diabetes [111]. Multiple studies on monocytes and the serum of long-lived and elderly individuals have revealed age-related miRNAs [112–114].

Monitoring the changes in miRNA expression during aging may be useful in detecting promising biomarkers [92]. In sarcopenia, biomarkers such as miR-181a, miR-434-3p, miR-431, miR-29, and miR-126 are involved in IGF-1, senescence, and apoptosis signaling in cells [115]. MiR-19a-3p has been recommended as a biomarker for ischemic stroke, and the gene pathways targeted by miRNAs related to inflammation, coagulation, and platelet activation have been identified [116]. Considering the association between stroke and age and that the elderly population has a higher risk of stroke, the identification of miRNAs can be used for various age-related diseases to subsequently discover biomarkers for disease treatment and prevention [117]. Similarly, human hearing loss is related to the expression of miR-34a and miR-21, which may be potential biomarkers of inflammation [118,119]. MiR455-3p has been proposed as a potential peripheral biomarker for Alzheimer's disease [120,121]. To elucidate the interaction among miRNA, aging diseases, the aging process, and the underlying mechanisms, there is need for more longitudinal studies and the integration into multi-omics methods.

LncRNAs are ≥200 nucleotides in length that are another type of noncoding RNA [122] and act as signals, baits, and guides during transcription and affect gene expression on different levels, including recombination, transcription regulation, and post-transcriptional modification [123], thereby affecting the length of life and aging. The downregulation of lncRNA induces decreased cell growth and senescence [94]. Telomere-lncRNA can regulate cell telomerase activity during aging [124]. Age-related lncRNA expression disorders may

affect neurogenesis and synaptic plasticity by promoting neuropathy via protein aggregation and neurodegeneration [124]. Meg3 has been thought to be related to cardiovascular diseases of aging [125].

CircRNAs are RNA transcripts produced by the reverse splicing of protein-coding exons. These transcripts may serve as useful biomarkers as they accumulate in the brain during aging [126]. CircRNAs can be detected in the blood [127,128], serum [129], and saliva [130]; they are very valuable biomarkers of aging [131,132]. A recent study has shown that, in multiple system atrophy (MSA) patients, circRNAs are upregulated [133]. In Table 2, we summarize the main potential aging biomarkers identified in transcriptomics studies.

Table 2. Potential aging biomarkers identified in transcriptomics studies.

Omics	Biomarkers	Function/Application	References
	Transcriptomics aging clocks	Predictors of age	[35]
	Transcriptome aging of skin fibroblasts	Determining the biological age	[36]
	The number of ABCG1	Determines human lifespan	[96,97]
	BIRC2 gene	An apoptosis regulator of inflammation, cell proliferation and mitotic kinase signal transduction	[99]
	The expression of 11 genes (*AMZ1, MANEAL, PARP3, KIAA0408, ISM1, CRIP1, NEFL, PHLDA3, DDB2, CHN1, CAPN2*)	Positively correlated with aging	[100]
	The expression of 4 genes (*MXRA8, SLC4A10, CD248,* and *PLEKHA7*)	Negatively correlated with aging	[100]
	miR-22-3p and miR-28-3p	Positively correlated with age	[92]
	miR-425-3p, miR-182-5p, miR-99b-5p, etc.	Negatively correlated with age	[92]
	miR-181a, miR-434-3p, miR-431, miR-29, and miR-126	In sarcopenia	[115]
Transcriptomics	miR-19a-3p	A biomarker for ischemic stroke	[116]
	the expression of miR-34a	Associated with human hearing loss	[118]
	miR-21	A potential biomarker of inflammation	[119]
	miR455-3p	As early biomarkers of AD	[120,121]
	lncRNAs	Provide different regulatory layers in the cell aging process, which can be used to intervene in this process	[124]
	Downregulation of lncRNA	Lung adenocarcinoma transcript 1 associated with metastasis in proliferating cells induces decreased cell growth	[94]
	Telomere-lncRNA	Can regulate the telomerase activity and survival rate of neural stem cells during aging	[124]
	Age-related lncRNA expression disorders	May affect neurogenesis and synaptic plasticity processes	[124]
	Meg3	Related to cardiovascular aging	[125]
	CircRNAs	May be valuable biomarkers in the aging brain	[126]
	Multiple circRNAs are upregulated	In multiple system atrophy (MSA), which is a sporadic neurodegenerative disease	[133]

4.3. Aging Proteomics

4.3.1. Proteomics-Based Biomarkers

Since it is closer to the phenotype than the genome or the transcriptome, the proteome has become an attractive target for researchers studying aging biomarkers. Proteins usually exert direct effects on information processing through signal transduction pathways. Based on a plasma soma scan, 1301 proteins from 120 participants were analyzed, and 10 major proteins related to aging were studied, i.e., GDF15, NPPB, CTSV, EFEMP1, ADAMTS5, CHRDL1, FSHB, MMP12, SOST, and PTN [43]. In a large cohort of 3301 patients, the proteomics aging clock was shown to accurately predict the age of a person [44]. Furthermore, urine tests are another way to use biomarkers to assess the rate of aging. For instance, urine collected from healthy people through the proteome is characterized by its higher abundance of LGGS3BP, MASP2, DNASE1, ANPEP, and IGFBP1 [134].

Proteomics can be used as an effective method to link the genotype and phenotype [135]. The relationship between circulating peptides (such as GDF8 and GDF11 pro-peptides and GDF8 and GDF11 mature proteins) and proteins has been implicated in accelerating aging phenotypes, and they are all involved in the inflammatory process [136]. The results are related to cardiovascular disease [137] and Alzheimer's disease [138] processes associated with proinflammatory cytokine profiles.

Therefore, proteomics is expected to decipher the aging process. The identified 11 differentially expressed proteins in the elderly may be useful biomarkers and could provide a basis for understanding the molecular mechanisms related to human health and aging [139]. In the plasma proteome of centenarians, the top 10 proteins related to nonhealthy aging (CRTAC1, CDKL1, CD14, and AOPEP) and healthy aging (TGFBI, TAS1R3, IGFAS, CRISP3, and CLEC3B) were revealed. These proteins can be used as aging biomarkers to develop new targets for clinical interventions [140]. The proteomic characteristics of 244 serum samples showed significant differences in the levels of 1312 proteins. Beneficial changes in human gene regulation can also be linked to longevity [141]. Proteomic studies have shown that serine protease inhibitors, SCT1, and GDF15 proteins can be used as biomarkers of aging, and there is an overlap in content considered as effective biomarkers [142]. In particular, GDF15, a mitogen involved in immune response and immune aging, is worthy of further study, because its concentration increases in elderly people regardless of gender or race [143].

Although some achievements have been made in aging proteomics, reliable proteomic biomarkers are still lacking. The main reasons include the accessibility of mass spectrometry technology, sample selection, maintaining the uniformity of preprocessing procedures, and the lack of proteomic heterogeneity and nonspecific circumvention for different populations, genders, and disease processes.

4.3.2. Senescence-Associated Secretory Phenotype-Based Biomarkers

SASP was first proposed by Coppe and colleagues [144]. Senescent cells can produce and secrete some cytokines, including include growth factors, inflammatory factors, and immunomodulators, to positively or negatively affect the surrounding cells and microenvironment [145]. Severe DNA damage can cause continuous DNA damage response signals and trigger SASP [146]. The NF-κB signaling pathway plays an important role in regulating the expression of IL-6 and IL-8 [147]. The activity of NF-κB is enhanced by mTOR activation and p38 MAPK upregulation [148,149], resulting in a potent SASP.

Sirtuins alone or cooperatively participate in mitochondrial function, cell cycle regulation, inflammation, DNA damage repair, and other biological processes, thus affecting the genome stability, inflammation alleviation, metabolic homeostasis, lifespan, and health maintenance [150,151]. Another protein beneficial for longevity and metabolic regulation is AMP-activated protein kinase (AMPK), affecting animal and human lifespans and health [152]. Besides, the overexpression of deacetylase family genes (Sirtuins) extends the lifespans of yeasts, worms, and fruit flies [153]. According to a recent study, Sir2 prolongs the lifespan by maintaining gene silencing during aging [154]. As another example,

telomerase repeatedly adds telomere DNA to chromosome ends to counteract telomere shortening associated with the cell cycle [155]. Further, inhibition of the mammalian target of the rapamycin (mTOR) signaling pathway [156] and mTOR regulatory signals has been proposed as a major molecular mechanism that delays aging in various organisms, from yeasts to mammals [157]. Finally, methionine sulfoxide is considered to be a marker of biological aging [158]. Methionine sulfoxide reductase is a specific antioxidant enzyme that removes this modification of proteins and, at the same time, acts as a general cellular antioxidant to scavenge free radicals and protect the cell from biological oxidative stress [159]. In Table 3, we summarize the main potential biomarkers of aging identified in proteomics studies.

Table 3. Potential aging biomarkers identified in proteomics studies.

Omics	Biomarkers	Function/Application	References
Proteomics	Proteomics aging clocks	Accurately predict the age of a person	[44]
	GDF15, PTN, ADAMTS5, FSHB, SOST, CHRDL1, NPPB, EFEMP1, MMP12, and CTSV	Related to aging	[43]
	LGALS3BP, MASP2, DNASE1, ANPEP, IGFBP1, etc.	Assess the rate of aging	[134]
	Circulating peptides (GDF8 and GDF11 pro-peptides and GDF8 and GDF11 mature proteins) and proteins	Be related to the accelerated dominant aging phenotype, and they are all involved in the inflammatory process	[136]
	CLEC3B, CRISP3, IGFAS, TAS1R3, and TGFBI	Be related to healthy aging	[140]
	AOPEP, CD14, CDKL1, and CRTAC1	Be related to nonhealthy aging	[140]
	Serine protease inhibitors, SCT1, and GDF15	As biomarkers of aging	[142]
	GDF15	A promising biomarker of aging	[143]
	Sirtuins	Affecting genome stability, inflammation alleviation, metabolic homeostasis, lifespan, and health maintenance	[150,151]
	The NF-κB signaling pathway	Regulating the expression of IL-6 and IL-8	[147]
	AMP-activated protein kinase (AMPK)	Affecting animal and human lifespan and health	[152]
	Telomerase	Counteract telomere shortening associated with the cell cycle	[155]
	Methionine sulfoxide	A marker of biological aging	[158]
	Methionine sulfoxide reductase	Protect the cell from biological oxidative stress	[159]

4.4. Aging Metabolomics

The biomarkers of aging metabolomics are the most useful tools for estimating aging. Metabolomics yields a huge amount of information, especially when it is used in conjunction with other omics approaches. Biomarkers based on metabolomics can simultaneously become the driving forces and signs of aging, which can then reveal the metabolic pathways related to the lifespan [160]. Changes in the metabolic profiles related to age can be quantitatively analyzed, and the analytical techniques for metabolite detection are highly sensitive and specific [161].

Regarding their clinical application, the number of known metabolomics biomarkers of aging is limited [162]. The use of multi-omics aging pipelines with transcriptomics or other types of omics data makes the metabolome itself valuable [163]. It is necessary to mention the "endophenotype" here. The aging process may be accompanied by different genetic, environmental, and disease effects, which may affect the overall transcriptome, proteome, and metabolome profiles. The multi-omics method based on the endophenotype is a more flexible method to detect age-related metabolome changes [164,165].

In a metabolomics and epigenetic analysis investigating aging in the British population, significant multi-directional changes in several metabolic pathways, such as CoA catabolism, vitamin E metabolism, lysine metabolism, tryptophan metabolism, and tyrosine metabolism, were detected [166]. In another study, 2678 plasma metabolites in a cohort of 150 individuals (30–100 years old) were identified [167]. The levels of monoacylglycerides, diacylglycerols, and phosphoserine decreased with age. On the other hand, the product of proteolysis, i.e., L-γ-glutamyl-L-leucine, increased independently of gender during aging. However, the levels of 25-hydroxy-hexanoic acid, eicosapentaenoic acid, and phosphoserine showed a negative trend in the elderly. Therefore, the effect of aging on lipid distribution can be detected [167].

Various molecular mechanisms underpin the genetic factors involved in longevity. The main factors that affect longevity and aging are growth hormone (GH) and insulin/insulin-like growth factor (insulin/IGF-1) pathways [168] in various organisms, from yeasts to mammals (including humans) [169,170]. Nicotinamide adenine dinucleotide (NAD^+) plays a vital role in mitochondrial electron transport, and it decreases with age [171]. Dietary supplementation with NAD^+ can help maintain health and extend the lifespan of mice [172,173]. Nicotinamide ribose supplementation can induce muscle and intestinal stem cells in aging mice to rejuvenate [174,175]. However, its potential antiaging effect has to be balanced with the potential tumorigenesis risk [176].Chronic inflammation in tissues is another cause of aging. It is triggered by signaling pathways related to the activity of transcription factor NF-κB [177]. Inhibiting the activity of NF-κB extends the life of fruit flies and mice [178]. Further, the autophagy–lysosomal signaling pathway eliminates toxic and easily aggregating proteins to maintain the normal cell functions in nematodes [179], fruit flies [180], mice [181], and other model organisms, and even human cells, to extend their lifespan.

In addition, individuals with higher levels of advanced glycation end products (AGEs) suffer from oxidative damage, leading to immune aging [182,183]. There are nine differentially expressed metabolites in men and women, which may serve as biomarkers of the aging process [184]. Another study of 44,168 individuals (18–109 years old) from 12 cohorts revealed that the metabolic profiles of polyunsaturated fatty acids/total fatty acids, histidine, and leucine may be indirect predictors of long-term mortality in clinical trials [185]. However, there are many challenges in clinical metabolomics [186]. In Table 4, we summarize the main potential aging biomarkers identified in metabolomics studies.

4.5. Aging Microbiomics

The microbiota exists in all parts of the human body, including the gastrointestinal tract, skin, respiratory tract, and so on. However, its abundance varies depending on tissue and organ types. The human microbiota consists of trillions of coexisting microorganisms, including bacteria, protozoa, archaea, viruses, and fungi [187,188]. The temporal change in microbial diversity and composition [189,190] is essential to human development and health [191]. Many factors affect the diversity and stability of the human microbiome, such as diet, genetics, environment, and antibiotics [192]. During aging, changes in physiology, diet, medications, and lifestyle can lead to changes in the gut microbiota [193].

Table 4. Potential aging biomarkers identified in metabolomics studies.

Omics	Biomarkers	Function/Application	References
Metabolomics	CoA catabolism, vitamin E metabolism, lysine metabolism, tryptophan metabolism, tyrosine metabolism, etc.	Related to aging	[166]
	Monoacylglycerides, diacylglycerols and phosphoserine, etc.	Show a decreasing trend with age	[167]
	The product of proteolysis and L-γ-glutamyl-L-leucine	Increases independently of gender during aging	[167]
	25-hydroxy-hexanoic acid, eicosapentaenoic acid, phosphoserine, etc.	Show a negative trend in the elderly	[167]
	Nicotinamide adenine dinucleotide (NAD$^+$)	Plays a vital role in mitochondrial electron transport. can help maintain health and extend the life of mice	[171–173]
	Higher advanced glycation end products (AGEs) levels	Suffered from oxidative damage, leading to immune aging	[182,183]
	Metabolic profile (polyunsaturated fatty acids/total fatty acids, histidine, leucine, etc.)	May be an indirect predictor of mortality related to clinical trials and medical decision-making	[185]
	Inhibiting the activity of NF-κB	Extends the life of fruit fly and mouse	[177,178]
	The autophagy–lysosomal signaling pathway	Maintain the normal cell functions and extend the lifespan	[179–181]

With the rapid development of high-throughput sequencing technologies, several breakthroughs have been achieved by studies on the gut microbiota [194]. Generally, the diversity of the microbiome decreases with age, especially in the elderly [195–197]. For example, *Bifidobacterium,* which plays a role in maintaining human health, is an important part of the gut microbiota [197]. However, the abundances of *Bacteroides* and *Enterobacteriaceae* increased [198]. The long-term supplementation of *Bifidobacterium* can enhance the memory of aging rats [199]. The ratio between Firmicutes and Bacteroidetes can be an indicator of metabolic health and decreases with age [200]. According to studies using human and animal models, the composition of the gut microbiota is an important factor related to longevity [201,202]. Some recent studies have shown that the composition of the human gut microbiota is affected by host age, diet, and environment [203,204]. The relationship of the human gut microbiota with metabolic disorders, obesity, inflammatory bowel disease, and infections has also been investigated [205,206]. The gut microbiota can produce various short-chain fatty acids, essential amino acids, peptides, vitamins, and other organic compounds. These microorganisms are also involved in the digestion and absorption of the gastrointestinal tract and regulate immune, metabolism, and other related physiological processes [207,208].

Considering the abovementioned findings, the relationship between the gut microbiota and healthy aging has been investigated [209,210]. In the elderly, the gut microbiota is related to host immune homeostasis caused by inflammation [211], which may lead to diseases and debilitating processes [212,213]. Chronic inflammation, neurodegeneration, and metabolic syndrome are related to inflammatory aging [198,214]. Immune senescence is usually accompanied by upregulation of the inflammatory response. During aging, the continuous imbalance in the gut microbiota leads to an inflammatory response in the intestinal mucosa [215]. Among the elderly, a specific microbiota phenotype has been detected, and the number of bacteria with anti-inflammatory and immunomodulatory effects, including *Bacteroides, Ruminococcus, Faecalibacterium, Parabacteroides*, and *Clostridium,* is reduced, which may promote the development of common diseases and disorders related to aging. [216,217].

In one of our unpublished studies, using high-throughput sequencing tools (16S rRNA gene amplicons and metagenomics), we obtained the longest trajectory of metagenomic changes in the human gut microbiota with age and characterized the microbiome of Chinese centenarians. The gut microbiota of Chinese centenarians was more diverse than that of young people. We observed the enrichment of several potentially beneficial bacterial groups, including those that produce SCFAs, in centenarians. However, some OTUs associated with beneficial bacteria (*Faecalibacterium*) were reduced. Our data suggest that longer lifespans are linked to health-related probiotics in centenarians. The relative abundances of *Akkermansia*, *Lactobacillus,* and *Christensenellaceae* increased in the elderly and centenarian groups. These bacterial families promote immune regulation, defend against inflammation, and promote healthy metabolic homeostasis [218,219]; therefore, they could be markers of the ecosystem of long-lived people.

In addition, we identified the taxonomy of "longevity-related strains" at the genus level and accurately described the functional changes that occur with aging. To validate these metagenomics results, we conducted in vitro screening and in vivo mouse experiments. We observed high oxidoreductase activity in the microbiota of centenarians and concluded that longevity-related strains play an antioxidant role in humans, thus contributing to healthy aging and longevity. In another study on the microbiome of centenarians, decreased concentrations of *Blautia*, *Coprococcus*, *Roseburia*, and *Faecalibacterium* and high concentrations of *Christensenellaceae*, *Akkermansia*, and *Bifidobacterium,* which are associated with immunological and metabolic health, and a significant increase in concentrations of *Desulfovibrionaceae* and *Enterobacteriaceae* were linked to longevity [220].

Combining at least two omics methods (genomics, transcriptomics, proteomics, and metabolomics) in the study of the microbiome advances the discovery of microbiome biomarkers of aging. A study analyzing the metabolites in human blood samples showed that, with age, the bacteria associated with the metabolism of indole and tryptophan significantly decreased in the gut microbiome. However, the downregulation of tryptophan transport and metabolism is essential for cognitive function and T-cell differentiation [221–223]. Tryptophan plays a vital role in intestinal immune tolerance and maintaining the balance of the gut microbiota [224]. The enhancement of tryptophan metabolism has been positively correlated with age, which is consistent with the finding that the serum tryptophan level in the elderly is low [225] and, in patients with dementia, is also reduced [226].

In summary, microbiomics is another promising field for diagnosing senile diseases, discovering novel clinical interventions, and establishing biomarkers of aging. However, future studies need to consider the effect of other species, such as archaea, fungi, and viruses, as well as that of the environment and host. These factors play a vital role in the overall regulation. Since aging is a complex and dynamic process, useful biomarkers in one population may not be applicable to different, other populations. Therefore, longitudinal research cohort studies should be conducted. The standardization of sample collection, processing, and data analysis protocols should also be considered. In Table 5, we summarize the main potential biomarkers of aging identified in microbiomics studies.

4.6. Early Biomarkers of Aging

Early biomarkers of aging are key, because it is unlikely that even the best antiaging interventions will be effective in aged individuals, and it would be more important to intervene earlier in life. The gut microbiota may influence the physiological mechanisms of a wide range of age-related diseases and biological phenotypes. It is worth noting that, compared with most age-related disease onsets and age-driven health declines, strategies to repair or improve the dynamics of the gut microbial community at or before this life stage may be useful ways to explore the prevention of premature aging, for example, by supplementing probiotics, targeted dietary changes, or vaccines. In addition, some different taxa may require further research on the physiological aging potential of the microbiota in early to mid-adulthood. Future research will verify these findings through richer interference factor controls, larger sample sizes, longitudinal follow-ups, and direct

immunological measurements, which will further support the gut microbiota to help in the early detection and prevention of accelerated aging and age-related diseases.

Table 5. Potential aging biomarkers identified in microbiomics studies.

Omics	Biomarkers	Function/Application	References
Microbiomics	The abundance of *Bifidobacterium*, *Bacteroides*, *Lactobacillus*, *Ruminococcus*, and *Bacillus* decreased, while the number of *Streptococcus*, *Enterobacter*, *Clostridium*, and *Escherichia* increased	During the aging process	[49]
	The ratio of Firmicutes to Bacteroidetes	Can be used as a criterion for metabolic health, and the ratio will decrease with age	[200]
	Bacteroides, *Ruminococcus*, *Faecalibacterium*, *Coprococcus*, *Parabacteroides*, *Clostridium*, *Alistipes*, etc.	Bacteria with anti-inflammatory and immunomodulatory effects	[216,217]
	Christensenellaceae, along with *Akkermansia* and *Lactobacillus*	Promote immune regulation, defend against inflammation, and promote healthy metabolic homeostasis	[218,219]
	Christensenellaceae, *Akkermansia*, *Bifidobacterium*	Associated with immunological and metabolic health	[220]
	Decrease in *Blautia*, *Coprococcus*, *Roseburia*, and *Faecalibacterium* and significant increase in *Desulfovibrionaceae* and *Enterobacteriaceae*	Linked to longevity	[220]
	Akkermansia, *Lactobacillus,* and *Christensenellaceae*	Longevity-related strains play an antioxidant role in humans, which helps achieve healthy aging and longevity	In our study

5. Integromics and Systems Biology

To promote the multidimensional analysis of data, advanced omics technology is inseparable from advanced omics analytical tools. At present, large-scale, high-quality, and high-throughput data from various omics methods can be efficiently and independently analyzed. However, separate data analysis and interpretation ignore the correlation and biological interference between different omics levels. Therefore, the integration of single-omics methods is essential for an in-depth understanding of the aging process and its mechanism.

Integromics, the comprehensive analysis of different omics data, and systems biology have provided several breakthroughs in the study of aging and antiaging interventions. Together, they have emerged as a more complex statistical method and combine the experimental data obtained in multiple omics methods with computational models to provide a holistic view of the aging landscape [227]. Considering the complexity and heterogeneity of aging, integromics and systems biology not only provide static maps of molecules but are also used to characterize the mutual changes of molecules over time. This helps determine the optimal time point for aging biomarker measurements and specific antiaging drug treatments. Each omics-level biomarker candidate based on integromics and systems biology has biological relevance. Significant biomarker candidates can be preferentially used as biomarkers of aging in medicine and as new antiaging targets.

Currently, data dimensionality reduction and normalization methods, such as a multifactor analysis or partial least square regression analysis, which can identify the main sources of data differences, are used in aging research [228,229]. Similarly, the principal component analysis method decomposes the data into several factors to facilitate the identification of the factor that can best explain the differential phenotypes among aging patients. Other multivariate analysis methods are used to study the overall correlation of

multiple variables and, finally, determine the variables that are most likely to shed light on the biological characteristics of specific differences. However, the combined influence of multiple factors and the high variability of a single dataset may cause difficulties in identifying biologically relevant and unrelated molecules [229].

Since aging is a multifactor, complex disease in which multiple physiological processes are regulated simultaneously and change over time, a single regulation is not sufficient to alleviate or reverse this pathological process. Thus, the use of integromics and systems biology to identify biologically interconnected processes in aging, which can be simultaneously regulated by combination therapy, is essential. Integromics and systems biology can also play an important role in personalized therapy. In cancer, multi-omics methods have been used for disease identification and personalized treatment [230,231]. In autism spectrum disorders, along with integration with clinical data, they have been used to accelerate the development of precision medicine and personalized medicine [232]. In addition, building computer models that predict the occurrence of certain diseases based on multi-omics could compare the biomarkers and pathways shared between diseases, thereby paving the way for efficient drug use. In Table 6, we summarize the main potential biomarkers of aging identified in integromics and systems biology studies.

Table 6. Potential aging biomarkers identified in integromics and systems biology studies.

Omics	Biomarkers	Function/Application	References
Integromics and systems biology	The method of comprehensive analysis of different omics data	This method combines experimental data of multiple omics levels with computational models and analyzes them as a whole to identify valuable data	[227]
	Multi-factor analysis or partial least square regression analysis	Can identify the main sources of data differences	[228,229]
	Multi-omics methods	Used for disease identification and personalized treatment in cancer	[230,231]
	Multi-omics and integration with clinical data	Used as a way to accelerate precision medicine and personalized medicine	[232]

6. Conclusions and Prospects

Rapid advances in science and technology have accelerated the arrival of the "omics era", thereby enabling researchers to collect and integrate data at different molecular levels. The identification of biomarkers of aging and new targets for antiaging interventions is crucial in aging biology and geriatrics. The multi-level information obtained through multi-omics technology contributes to the increased understanding of the mechanisms of aging and provides new opportunities for the diagnosis and treatment of aging and aging-related diseases.

We have summarized the various omics techniques used to characterize aging biomarkers. Each screened biomarker is a promising candidate and can be integrated into an "aging biomarker library" that can serve as a diagnostic and prognostic tool. Here, we mainly categorized them based on the existing biomarkers of aging. We summarized the recent omics methods used to discover biomarkers in genomics, transcriptomics, proteomics, metabolomics, and metagenomics (Figure 1). In the field of geriatrics, discovering new biomarkers from existing datasets and new biological age measurement methods are of great value. At present, a more accurate biological age measurement method based on the aging clock of DNA methylation is needed, which can also be analyzed and evaluated through the transcriptome aging clock. The epigenetic clock from the comparative analysis of actual age and biological age shows that the aging process is inherently related to biological age. In addition, biological age can be measured using transcription profiles. MiRNAs, lncRNAs, and circRNAs contribute to the discovery of novel biomarkers of aging. Proteomics is receiving increasing attention in aging research, because their findings are

the most reproducible and the easiest to verify. However, the application of proteomics technology has some limitations, such as high costs and a lack of accurate clinical practice applications. The biomarkers identified in metabolomics and microbiomics studies also have great potential; however, their application in clinical practice is limited by the limited number of longitudinal metabolomics studies available. The comparison of methods from single-omics is the key aspect to better illustrate how integrating these methods will help. This will serve as a synthesis of information rather than just data management. For example, we can compare and analyze the genomics, transcriptomics, proteomics, and metabolomics in microbiology and the substances that appear in the analysis of genomics, transcriptomics, proteomics, and metabolomics in longitudinal populations. For example, we analyzed the correlation between the properties of metabolites regulated by the gut microbiome of healthy and long-lived people and the biomarkers analyzed from blood metabolomics of healthy and long-lived people. This was a good way to show that the integration of what we learned from each method was not just the sum of its parts.
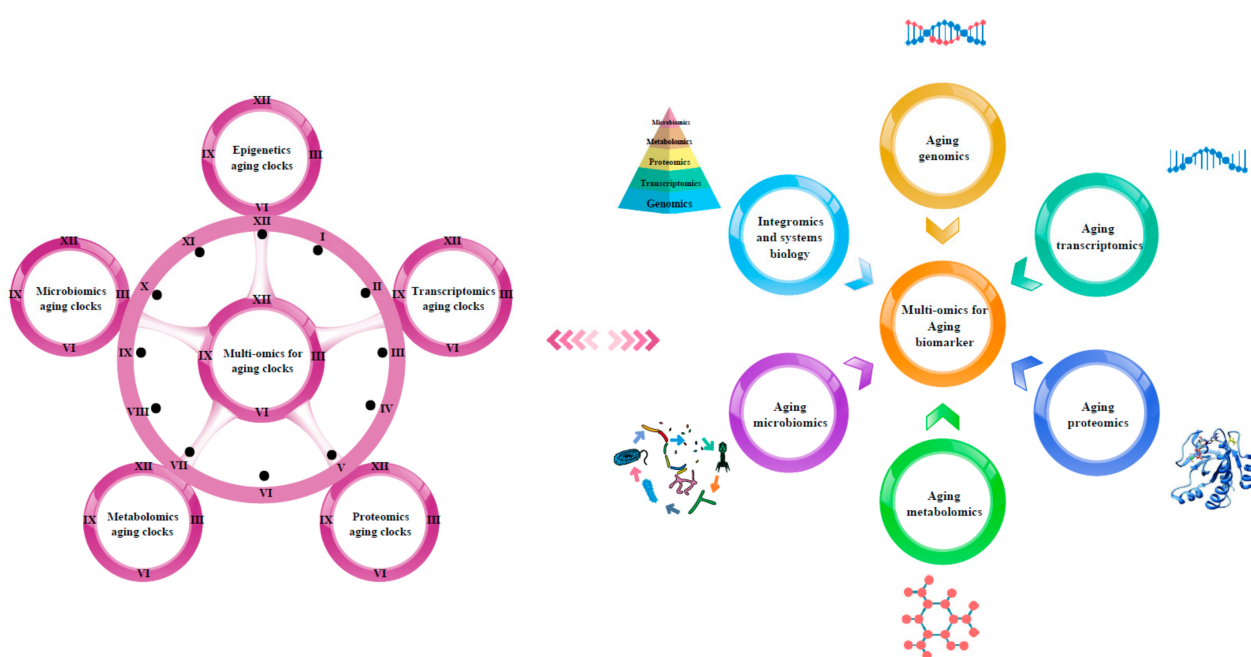
Figure 1. Multi-omics-based technologies for characterizing aging clocks and biomarkers. Aging is a comprehensive process affected by multiple factors that is associated with changes at the molecular, cellular, tissue, and organism levels, thus requiring objective analytical research tools. The integrated multi-omics approach is essential to achieve a comprehensive understanding of the biological mechanisms of aging.

In the context of personalized and precision medicine, multi-omics methods have attracted widespread attention, because they can provide an in-depth understanding of the molecular patterns and cover a wide range of characteristics, such as participating in the metabolic, genetic, and signal transduction pathways of complex aging [233]. Therefore, we suggest that a combination of multiple biomarkers for a comprehensive diagnosis and systematic analysis can objectively characterize the aging process (Figure 2). Integromics and systems biology methodologies can provide insights into organ- and system-specific functions; reflect the phenotype and the processes involved in metabolism, immunity, and structure; and function in different physiological domains and their rates of change in an individual's lifetime [234]. It can correlate the results at different levels of complexity with clinical profiles. Aging clocks and aging biomarkers and their combinations with multi-omics are usually investigated in experimental studies. However, due to their practicality and feasibility, they are becoming more popular topics in clinical medical research, which

advances the knowledge on human aging. Biomarkers have great application prospects in drug target screening, because biomarkers and targets can be the same substance, and they have a high potential for mutual transformation [235]. Methylated aging can evaluate antiaging interventions to develop new types of aging clocks [236]. Biological age is a predictor of mortality in ischemic stroke [237]. In addition, the biological age of the brain can serve as a prognostic tool [238]. Biomarkers are also used for drug discovery and utilization. Designing studies based on biomarkers can help eliminate hidden errors in the treatment process [239]. Recently, a novel biomarker-based miRNA therapeutic strategy for hepatocellular carcinoma (HCC) was successfully applied [240]. Using noninvasive biomarker-based methods, biomarkers contribute to a better understanding of the patho-physiological mechanism of diseases [241]. This review not only focused on single-omics methods to characterize aging biomarkers but, more importantly, integrated multiple omics to evaluate the relevance of these biomarkers and maximized a systematic analysis of the data.
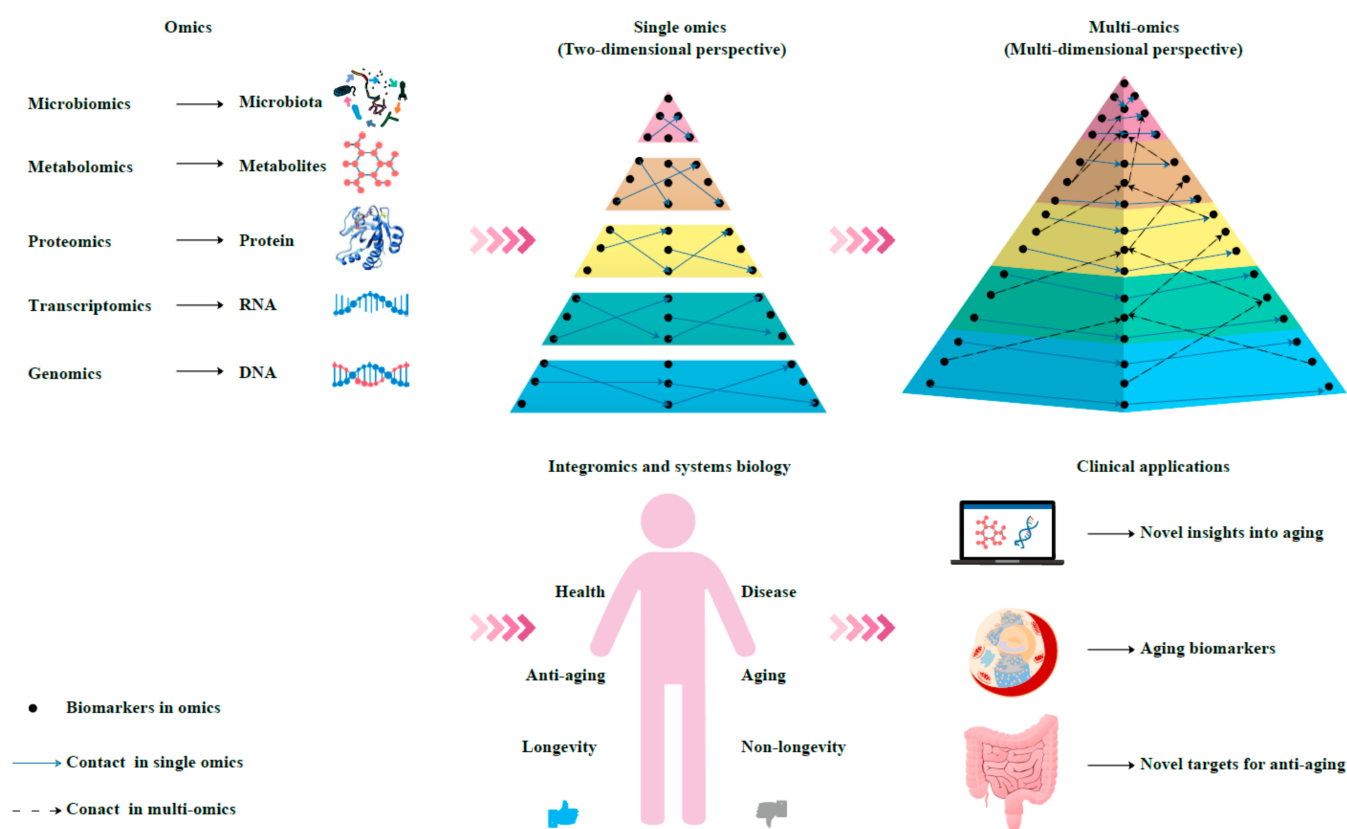
Figure 2. Schematic diagram of an integrated multi-omics approach to the research and application of aging biomarkers. Genomics, transcriptomics, proteomics, metabolomics, and microbiomics enable the high-throughput quantitative profiling of molecules in biological systems to reveal aging-related changes. Combining single-omics data with integromics and systems biology contributes to an increased understanding of the mechanisms of aging and paves the way for the development and utilization of aging biomarkers and novel antiaging targets.

Although multi-omics methods have great potential, limitations and challenges remain. First, omics methods are expensive and require special equipment and highly qualified data analysis personnel. Second, the data quality can be uneven, the data source can be inaccurate, and nonstandard sampling can cause problems in data collection and verification. To date, research platforms and bioinformatics methods for processing large-scale omics data have not yet been standardized. For example, the biggest challenge in metabolomics studies lies in data processing and analysis due to the thousands of metabolites present

in biological organisms. Furthermore, the differences among individuals and inconsistent data processing and analysis programs add to this difficulty. Therefore, it is necessary to promote cross-disciplinary efforts and the standardization of procedures to increase the relevance of metabolomics-based strategies in clinical research. Longitudinal cohort studies with large samples should also be conducted. In addition, there is a lack of longitudinal samples and longitudinal studies. In order to delineate the sequence of events, a longitudinal study of the microbiota with age is required. Model organisms with short lifespans and less complex microbiota and established biomarkers of aging make this easier to study. For example, in nematodes, fruit flies, and mice, the integrity of the intestinal epithelium/barrier has been shown to decline with age and is also associated with human aging. Since aging is a complex process that occurs at all levels of biological systems, the impact of antiaging interventions on clinical practice requires a multi-dimensional and systematic approach.

Cellular aging, leading to tissue dysfunction, is widely accepted as contributing to aging and the development of debilitating age-related diseases. Senolytics and bioflavonoids are the key in anti-aging research. Endogenous defenses against ROS include the enzymes superoxide dismutase, glutathione peroxidase, catalase, and peroxiredoxins and the nonenzymatic antioxidants, glutathione, thioredoxin, and uric acid. There are many nonenzymatic endogenous antioxidants. Cofactor coenzyme Q is present in cells and membranes and plays an important role in cellular metabolism and in the respiratory chain. Vitamin A combines with peroxyl radicals, thus preventing lipid peroxidation. Uric acid prevents the lysis of erythrocytes and is also an important scavenger of singlet oxygen. Other small molecular weight nonenzymatic antioxidants include vitamins E and C and many minerals like selenium and zinc. Selenium is the integral part of the antioxidant enzyme glutathione peroxidase. Flavonoids (i.e., flavonols, flavanols, anthocyanins, isoflavonoids, flavonones, flavones, and phenolic acids) act as chelators of transition metal ions involved in Fenton chemistry and ROS scavengers. Bioflavonoids can adjust blood lipids, extend the life of red blood cells, effectively remove free radicals and toxins in the body, and prevent and reduce the occurrence of diseases. Modern pharmacological research shows that curcumin has anti-inflammatory, antioxidant, antitumor, and other pharmacological effects. Resveratrol is a natural antioxidant found in plants. Quercetin may reduce the consumption of glutathione, increase the activity of antioxidant enzymes, and directly or indirectly exert an antioxidant effect in the body after being absorbed by the intestine.

Advances in computer science, including meta-analysis and artificial intelligence, are expected to remarkably increase the speed and efficiency of aging biomarker research [242]. However, before their application in the clinical setting, candidate biomarkers should be verified. This verification process must include larger sample populations. Despite the large gap between the identification of useful biomarkers and their application in clinical practice, the integrated analysis of multi-omics data is a promising tool to identify new candidate biomarkers that could be developed and used to identify pharmaceutical targets and improve human health during aging, thereby advancing our understanding of the pathophysiology of the complex and dynamic process of aging.

Author Contributions: L.W.: Data curation, Conceptualization, Methodology, Formal analysis, Investigation, Writing—original draft, Writing—review and editing, and Visualization. X.X.: Conceptualization, Methodology, Formal analysis, and Investigation. T.L.: Conceptualization, Methodology, and Investigation. J.M.: Conceptualization, Methodology, Formal analysis, Investigation, and Visualization. L.Y.: Methodology and Investigation. J.Y.: Methodology and Investigation. L.L.: Methodology and Investigation. Y.X.: Methodology and Investigation. H.L.: Methodology and Investigation. J.Z.: Conceptualization, Methodology, Resources, Writing—review and editing, and Funding acquisition. X.C.: Methodology and Investigation. Y.D.: Conceptualization, Methodology, Formal analysis, Writing—review and editing, Supervision, and Validation. Q.W.: Conceptualization, Methodology, Resources, Writing—review and editing, Funding acquisition, Supervision, and Validation. All authors have read and agreed to the published version of the manuscript.

References

1. United Nations, Department of Economic and Social Affairs, Population Division. *World Population Prospects 2019: Highlights*; ST/ESA/SER.A/423; United Nation: New York, NY, USA, 2019. Available online: https://population.un.org/wpp/Publications/Files/WPP2019_Highlights.pdf (accessed on 21 January 2021).
2. Carmona, J.J.; Michan, S. Biology of healthy aging and longevity. *Rev. Investig. Clin.* **2016**, *68*, 7–16.
3. Tuttle, C.S.L.; Waaijer, M.E.C.; Slee-Valentijn, M.S.; Stijnen, T.; Westendorp, R.; Maier, A.B. Cellular senescence and chronological age in various human tissues: A systematic review and meta-analysis. *Aging Cell* **2020**, *19*, e13083. [CrossRef] [PubMed]
4. Denic, A.; Glassock, R.J.; Rule, A.D. Structural and functional changes with the aging kidney. *Adv. Chronic Kidney Dis.* **2016**, *23*, 19–28. [CrossRef]
5. Kim, I.H.; Kisseleva, T.; Brenner, D.A. Aging and liver disease. *Curr. Opin. Gastroenterol.* **2015**, *31*, 184–191. [CrossRef]
6. Bernardes de Jesus, B.; Blasco, M.A. Potential of telomerase activation in extending health span and longevity. *Curr. Opin. Cell Biol.* **2012**, *24*, 739–743. [CrossRef]
7. Gross, A.L.; Carlson, M.C.; Chu, N.M.; McAdams-DeMarco, M.A.; Mungas, D.; Simonsick, E.M. Derivation of a measure of physiological multisystem dysregulation: Results from WHAS and health ABC. *Mech. Ageing Dev.* **2020**, *188*, 111258. [CrossRef] [PubMed]
8. Belloni, G.; Cesari, M. Frailty and intrinsic capacity: Two distinct but related constructs. *Front. Med.* **2019**, *6*, 133. [CrossRef] [PubMed]
9. Zhavoronkov, A.; Mamoshina, P.; Vanhaelen, Q.; Scheibye-Knudsen, M.; Moskalev, A.; Aliper, A. Artificial intelligence for aging and longevity research: Recent advances and perspectives. *Ageing Res. Rev.* **2019**, *49*, 49–66. [CrossRef]
10. Buerkle, A.; Moreno-Villanueva, M.; Bernhard, J.; Blasco, M.; Zondag, G.; Hoeijmakers, J.H.J.; Toussaint, O.; Grubeck-Loebenstein, B.; Mocchegiani, E.; Collino, S.; et al. MARK-AGE biomarkers of ageing. *Mech. Ageing Dev.* **2015**, *151*, 2–12. [CrossRef]
11. Pilling, L.C.; Atkins, J.L.; Bowman, K.; Jones, S.E.; Tyrrell, J.; Beaumont, R.N.; Ruth, K.S.; Tuke, M.A.; Yaghootkar, H.; Wood, A.R.; et al. Human longevity is influenced by many genetic variants: Evidence from 75,000 UK Biobank participants. *Aging* **2016**, *8*, 547–563. [CrossRef]
12. Zhavoronkov, A.; Buzdin, A.A.; Garazha, A.V.; Borisov, N.M.; Moskalev, A.A. Signaling pathway cloud regulation for in silico screening and ranking of the potential geroprotective drugs. *Front. Genet.* **2014**, *5*, 49. [CrossRef]
13. Gott, A.; Andrews, C.; Hormigos, M.L.; Spencer, K.; Bateson, M.; Nettle, D. Chronological age, biological age, and individual variation in the stress response in the European starling: A follow-up study. *PeerJ* **2018**, *6*, e5842. [CrossRef] [PubMed]
14. Brown, P.J.; Wall, M.M.; Chen, C.; Levine, M.E.; Yaffe, K.; Roose, S.P.; Rutherford, B.R. Biological age, not chronological age, is associated with late-life depression. *J. Gerontol. Ser. A Biol. Sci. Med. Sci.* **2018**, *73*, 1370–1376. [CrossRef] [PubMed]
15. Kim, S.J.; Kim, B.J.; Kang, H. Measurement of biological age may help to assess the risk of colorectal adenoma in screening colonoscopy. *World J. Gastroenterol.* **2017**, *23*, 6877–6883. [CrossRef]
16. Cho, I.H.; Park, K.S.; Lim, C.J. An empirical comparative study on biological age estimation algorithms with an application of Work Ability Index (WAI). *Mech. Ageing Dev.* **2010**, *131*, 69–78. [CrossRef]
17. Klemera, P.; Doubal, S. A new approach to the concept and computation of biological age. *Mech. Ageing Dev.* **2006**, *127*, 240–248. [CrossRef] [PubMed]
18. Bai, X.J.; Han, L.L.; Liu, Q.; Shan, H.Y.; Lin, H.L.; Sun, X.F.; Chen, X.M. Evaluation of biological aging process—A population-based study of healthy people in china. *Gerontology* **2010**, *56*, 129–140. [CrossRef]
19. Zhang, W.G.; Bai, X.J.; Sun, X.F.; Cai, G.Y.; Bai, X.Y.; Zhu, S.Y.; Zhang, M.; Chen, X.M. Construction of an integral formula of biological age for a healthy Chinese population using principle component analysis. *J. Nutr. Health Aging* **2014**, *18*, 137–142. [CrossRef] [PubMed]
20. Belsky, D.W.; Caspi, A.; Houts, R.; Cohen, H.J.; Corcoran, D.L.; Danese, A.; Harrington, H.; Israel, S.; Levine, M.E.; Schaefer, J.D. Quantification of biological aging in young adults. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, E4104–E4110. [CrossRef] [PubMed]
21. Mitnitski, A.; Howlett, S.E.; Rockwood, K. Heterogeneity of human aging and its assessment. *J. Gerontol. Ser. A Biol. Sci. Med. Sci.* **2017**, *72*, 877–884. [CrossRef]
22. Horvath, S.; Raj, K. DNA methylation-based biomarkers and the epigenetic clock theory of ageing. *Nat. Rev. Genet.* **2018**, *19*, 371–384. [CrossRef] [PubMed]

23. Levine, M.E.; Lu, A.T.; Quach, A.; Chen, B.H.; Assimes, T.L.; Bandinelli, S.; Hou, L.; Baccarelli, A.A.; Stewart, J.D.; Li, Y.; et al. An epigenetic biomarker of aging for lifespan and healthspan. *Aging* **2018**, *10*, 573–591. [CrossRef]

24. Hannum, G.; Guinney, J.; Zhao, L.; Zhang, L.; Hughes, G.; Sadda, S.; Klotzle, B.; Bibikova, M.; Fan, J.B.; Gao, Y.; et al. Genome-wide methylation profiles reveal quantitative views of human aging rates. *Mol. Cell* **2013**, *49*, 359–367. [CrossRef] [PubMed]

25. Liu, Z.; Leung, D.; Levine, M. Comparative analysis of epigenetic aging clocks from CpG characteristics to functional associations. *bioRxiv* **2019**, *51*, 512483. [CrossRef]

26. Lin, Q.; Weidner, C.I.; Costa, I.G.; Marioni, R.E.; Ferreira, M.R.P.; Deary, I.J.; Wagner, W. DNA methylation levels at individual age-associated CpG sites can be indicative for life expectancy. *Aging* **2016**, *8*, 394–401. [CrossRef] [PubMed]

27. Yang, Z.; Wong, A.; Kuh, D.; Paul, D.S.; Rakyan, V.K.; Leslie, R.D.; Zheng, S.J.C.; Widschwendter, M.; Beck, S.; Teschendorff, A. Correlation of an epigenetic mitotic clock with cancer risk. *Genome Biol.* **2016**, *17*, 1–8. [CrossRef]

28. Zhang, Y.; Wilson, R.; Heiss, J.; Breitling, L.P.; Saum, K.U.; Schoettker, B.; Holleczek, B.; Waldenberger, M.; Peters, A.; Brenner, H. DNA methylation signatures in peripheral blood strongly predict all-cause mortality. *Nat. Commun.* **2017**, *8*, 14617. [CrossRef]

29. Weidner, C.I.; Lin, Q.; Koch, C.M.; Eisele, L.; Beier, F.; Ziegler, P.; Bauerschlag, D.O.; Joeckel, K.H.; Erbel, R.; Muehleisen, T.W.; et al. Aging of blood can be tracked by DNA methylation changes at just three CpG sites. *Genome Biol.* **2014**, *15*, R24. [CrossRef] [PubMed]

30. Garagnani, P.; Bacalini, M.G.; Pirazzini, C.; Gori, D.; Giuliani, C.; Mari, D.; Di Blasio, A.M.; Gentilini, D.; Vitale, G.; Collino, S.; et al. Methylation of ELOVL2 gene as a new epigenetic marker of age. *Aging Cell* **2012**, *11*, 1132–1134. [CrossRef]

31. Horvath, S.; Oshima, J.; Martin, G.M.; Lu, A.T.; Quach, A.; Cohen, H.; Felton, S.; Matsuyama, M.; Lowe, D.; Kabacik, S.; et al. Epigenetic clock for skin and blood cells applied to Hutchinson Gilford Progeria Syndrome and ex vivo studies. *Aging* **2018**, *10*, 1758–1775. [CrossRef] [PubMed]

32. Bocklandt, S.; Lin, W.; Sehl, M.E.; Sanchez, F.J.; Sinsheimer, J.S.; Horvath, S.; Vilain, E. Epigenetic Predictor of Age. *PLoS ONE* **2011**, *6*, e14821. [CrossRef]

33. Lu, A.T.; Quach, A.; Wilson, J.G.; Reiner, A.P.; Aviv, A.; Raj, K.; Hou, L.F.; Baccarelli, A.A.; Li, Y.; Stewart, J.D.; et al. DNA methylation GrimAge strongly predicts lifespan and healthspan. *Aging* **2019**, *11*, 303–327. [CrossRef] [PubMed]

34. Wang, M.; Lemos, B. Ribosomal DNA harbors an evolutionarily conserved clock of biological aging. *Genome Res.* **2019**, *29*, 325–333. [CrossRef] [PubMed]

35. Peters, M.J.; Joehanes, R.; Pilling, L.C.; Schurmann, C.; Conneely, K.N.; Powell, J.; Reinmaa, E.; Sutphin, G.L.; Zhernakova, A.; Schramm, K.; et al. The transcriptional landscape of age in human peripheral blood. *Nat. Commun.* **2015**, *6*, 9570. [CrossRef]

36. Fleischer, J.G.; Schulte, R.; Tsai, H.H.; Tyagi, S.; Ibarra, A.; Shokhirev, M.N.; Huang, L.; Hetzer, M.; Navlakha, S. Predicting age from the transcriptome of human dermal fibroblasts. *Genome Biol.* **2018**, *19*, 221. [CrossRef] [PubMed]

37. Putin, E.; Mamoshina, P.; Aliper, A.; Korzinkin, M.; Moskalev, A.; Kolosov, A.; Ostrovskiy, A.; Cantor, C.; Vijg, J.; Zhavoronkov, A. Deep biomarkers of human aging: Application of deep neural networks to biomarker development. *Aging* **2016**, *8*, 1021–1033. [CrossRef]

38. Horvath, S. DNA methylation age of human tissues and cell types. *Genome Biol.* **2013**, *14*, 1168. [CrossRef]

39. Mamoshina, P.; Kochetov, K.; Putin, E.; Aliper, A.; Zhavoronkov, A. Testing for batch effect through age predictors. *bioRxiv* **2019**, 531863. [CrossRef]

40. Bathke, J.; Konzer, A.; Remes, B.; McIntosh, M.; Klug, G. Comparative analyses of the variation of the transcriptome and proteome of Rhodobacter sphaeroides throughout growth. *BMC Genom.* **2019**, *20*, 358. [CrossRef]

41. Haider, S.; Pal, R. Integrated analysis of transcriptomic and proteomic data. *Curr. Genom.* **2013**, *14*, 91–110. [CrossRef]

42. Tin, A.; Yu, B.; Ma, J.; Masushita, K.; Daya, N.; Hoogeveen, R.C.; Ballantyne, C.M.; Couper, D.; Rebholz, C.M.; Grams, M.E.; et al. Reproducibility and variability of protein analytes measured using a multiplexed modified aptamer assay. *J. Appl. Lab. Med.* **2019**, *4*, 30–39. [CrossRef] [PubMed]

43. Tanaka, T.; Biancotto, A.; Moaddel, R.; Moore, A.Z.; Gonzalez-Freire, M.; Aon, M.A.; Candia, J.; Zhang, P.B.; Cheung, F.; Fantoni, G.; et al. Plasma proteomic signature of age in healthy humans. *Aging Cell* **2018**, *17*, e12799. [CrossRef]

44. Johnson, A.A.; Shokhirev, M.N.; Wyss-Coray, T.; Lehallier, B. Systematic review and analysis of human proteomics aging studies unveils a novel proteomic aging clock and identifies key processes that change with age. *Ageing Res. Rev.* **2020**, *60*, 101070. [CrossRef]

45. Lehallier, B.; Gate, D.; Schaum, N.; Nanasi, T.; Lee, S.E.; Yousef, H.; Losada, P.M.; Berdnik, D.; Keller, A.; Verghese, J.; et al. Undulating changes in human plasma proteome profiles across the lifespan. *Nat. Med.* **2019**, *25*, 1843–1850. [CrossRef] [PubMed]

46. Hertel, J.; Friedrich, N.; Wittfeld, K.; Pietzner, M.; Budde, K.; Van der Auwera, S.; Lohmann, T.; Teumer, A.; Voelzke, H.; Nauck, M.; et al. Measuring biological age via metabonomics: The metabolic age score. *J. Proteome Res.* **2016**, *15*, 400–410. [CrossRef] [PubMed]

47. van den Akker, E.B.; Trompet, S.; Wolf, J.J.H.B.; Beekman, M.; Suchiman, H.E.D.; Deelen, J. Predicting biological age based on the BBMRI-NL 1H-NMR metabolomics repository. *bioRxiv* **2019**, 632919. [CrossRef]

48. Choi, J.; Hur, T.Y.; Hong, Y. Influence of altered gut microbiota composition on aging and aging-related diseases. *J. Lifestyle Med.* **2018**, *8*, 1–7. [CrossRef]

49. Galkin, F.; Mamoshina, P.; Aliper, A.; Putin, E.; Moskalev, V.; Gladyshev, V.N.; Zhavoronkov, A. Human gut microbiome aging clock based on taxonomic profiling and deep learning. *iScience* **2020**, *23*, 101199. [CrossRef]

50. Claesson, M.J.; Jeffery, I.B.; Conde, S.; Power, S.E.; O'Connor, E.M.; Cusack, S.; Harris, H.M.B.; Coakley, M.; Lakshminarayanan, B.; O'Sullivan, O.; et al. Gut microbiota composition correlates with diet and health in the elderly. *Nature* **2012**, *488*, 178–184. [CrossRef]

51. Woodmansey, E.J.; McMurdo, M.E.T.; Macfarlane, G.T.; Macfarlane, S. Comparison of compositions and metabolic activities of fecal microbiotas in young adults and in antibiotic-treated and non-antibiotic-treated elderly subjects. *Appl. Environ. Microbiol.* **2004**, *70*, 6113–6122. [CrossRef]

52. Gonzalez-Bautista, E.; Andrieu, S.; Gutierrez-Robledo, L.M.; Garcia-Chanes, R.E.; De Souto Barreto, P. In the quest of a standard index of intrinsic capacity. A critical literature review. *J. Nutr. Health Aging* **2020**, *24*, 959–965. [CrossRef]

53. Buck Louis, G.M.; Smarr, M.M.; Patel, C.J. The exposome research paradigm: An opportunity to understand the environmental basis for human health and disease. *Curr. Environ. Health Rep.* **2017**, *4*, 89–98. [CrossRef]

54. Horvath, S.; Gurven, M.; Levine, M.E.; Trumble, B.C.; Kaplan, H.; Allayee, H.; Ritz, B.R.; Chen, B.; Lu, A.T.; Rickabaugh, T.M.; et al. An epigenetic clock analysis of race/ethnicity, sex, and coronary heart disease. *Genome Biol.* **2016**, *17*, 171. [CrossRef] [PubMed]

55. Day, K.; Waite, L.L.; Thalacker-Mercer, A.; West, A.; Bamman, M.M.; Brooks, J.D.; Myers, R.M.; Absher, D. Differential DNA methylation with age displays both common and dynamic features across human tissues that are influenced by CpG landscape. *Genome Biol.* **2013**, *14*, R102. [CrossRef]

56. Horvath, S.; Zhang, Y.; Langfelder, P.; Kahn, R.S.; Boks, M.P.M.; van Eijk, K.; van den Berg, L.H.; Ophoff, R. Aging effects on DNA methylation modules in human brain and blood tissue. *Genome Biol.* **2012**, *13*, R97. [CrossRef] [PubMed]

57. Horvath, S.; Ritz, B.R. Increased epigenetic age and granulocyte counts in the blood of Parkinson's disease patients. *Aging* **2015**, *7*, 1130–1142. [CrossRef] [PubMed]

58. Levine, M.E.; Hosgood, H.D.; Chen, B.; Absher, D.; Assimes, T.; Horvath, S. DNA methylation age of blood predicts future onset of lung cancer in the women's health initiative. *Aging* **2015**, *7*, 690–700. [CrossRef]

59. Vidal-Bralo, L.; Lopez-Golan, Y.; Gonzalez, A. Simplified assay for epigenetic age estimation in whole blood of adults. *Front. Genet.* **2016**, *7*, 126. [CrossRef]

60. Seaborne, R.A.; Strauss, J.; Cocks, M.; Shepherd, S.; O'Brien, T.D.; van Someren, K.A.; Bell, P.G.; Murgatroyd, C.; Morton, J.P.; Stewart, C.E.; et al. Human skeletal muscle possesses an epigenetic memory of hypertrophy. *Sci. Rep.* **2018**, *8*, 1898. [CrossRef]

61. Quach, A.; Levine, M.E.; Tanaka, T.; Lu, A.T.; Chen, B.H.; Ferrucci, L.; Ritz, B.; Bandinelli, S.; Neuhouser, M.; Beasley, J.; et al. Epigenetic clock analysis of diet, exercise, education, and lifestyle factors. *Aging* **2017**, *9*, 419–446. [CrossRef]

62. Giannakou, M.E.; Goss, M.; Junger, M.A.; Hafen, E.; Leevers, S.J.; Partridge, L. Long-lived Drosophila with overexpressed dFOXO in adult fat body. *Science* **2004**, *305*, 361. [CrossRef]

63. Bluher, M.; Kahn, B.B.; Kahn, C.R. Extended longevity in mice lacking the insulin receptor in adipose tissue. *Science* **2003**, *299*, 572–574. [CrossRef] [PubMed]

64. Tan, Q.; Soerensen, M.; Kruse, T.A.; Christensen, K.; Christiansen, L. A novel permutation test for case-only analysis identifies epistatic effects on human longevity in the FOXO gene family. *Aging Cell* **2013**, *12*, 690–694. [CrossRef]

65. Mahley, R.W.; Huang, Y. Apolipoprotein E sets the stage: Response to injury triggers neuropathology. *Neuron* **2012**, *76*, 871–885. [CrossRef] [PubMed]

66. Tyner, S.D.; Venkatachalam, S.; Choi, J.; Jones, S.; Ghebranious, N.; Igelmann, H.; Lu, X.B.; Soron, G.; Cooper, B.; Brayton, C.; et al. p53 mutant mice that display early ageing-associated phenotypes. *Nature* **2002**, *415*, 45–53. [CrossRef] [PubMed]

67. Blackburn, E.H. Telomeres and telomerase: Their mechanisms of action and the effects of altering their functions. *FEBS Lett.* **2005**, *579*, 859–862. [CrossRef]

68. Honig, L.S.; Kang, M.S.; Cheng, R.; Eckfeldt, J.H.; Thyagarajan, B.; Leiendecker-Foster, C.; Province, M.A.; Sanders, J.L.; Perls, T.; Christensen, K.; et al. Heritability of telomere length in a study of long-lived families. *Neurobiol. Aging* **2015**, *36*, 2785–2790. [CrossRef]

69. Blackburn, E.H.; Epel, E.S.; Lin, J. Human telomere biology: A contributory and interactive factor in aging, disease risks, and protection. *Science* **2015**, *350*, 1193–1198. [CrossRef]

70. Boccardi, V.; Paolisso, G. Telomerase activation: A potential key modulator for human healths pan and longevity. *Ageing Res. Rev.* **2014**, *15*, 1–5. [CrossRef] [PubMed]

71. Strong, M.A.; Vidal-Cardenas, S.L.; Karim, B.; Yu, H.; Guo, N.; Greider, C.W. Phenotypes in mTERT(+/−) and mTERT(−/−) mice are due to short telomeres, not telomere-independent functions of telomerase reverse transcriptase. *Mol. Cell. Biol.* **2011**, *31*, 2369–2379. [CrossRef]

72. Jaskelioff, M.; Muller, F.L.; Paik, J.H.; Thomas, E.; Jiang, S.; Adams, A.C.; Sahin, E.; Kost-Alimova, M.; Protopopov, A.; Cadinanos, J.; et al. Telomerase reactivation reverses tissue degeneration in aged telomerase-deficient mice. *Nature* **2011**, *469*, 102–106. [CrossRef]

73. Wang, Q.; Zhan, Y.; Pedersen, N.L.; Fang, F.; Hagg, S. Telomere length and all-cause mortality: A meta-analysis. *Ageing Res. Rev.* **2018**, *48*, 11–20. [CrossRef]

74. Lapham, K.; Kvale, M.N.; Lin, J.; Connell, S.; Croen, L.A.; Dispensa, B.P.; Fang, L.; Hesselson, S.; Hoffmann, T.J.; Iribarren, C.; et al. Automated assay of telomere length measurement and informatics for 100,000 subjects in the genetic epidemiology research on adult health and aging (GERA) cohort. *Genetics* **2015**, *200*, 1061–1072. [CrossRef]

75. Simoncini, T.; Hafezl-Moghadam, A.; Brazil, D.P.; Ley, K.; Chin, W.W.; Liao, J.K. Interaction of oestrogen receptor with the regulatory subunit of phosphatidylinositol-3-OH kinase. *Nature* **2000**, *407*, 538–541. [CrossRef] [PubMed]

76. Austad, S.N. Why women live longer than men: Sex differences in longevity. *Gend. Med.* **2006**, *3*, 79–92. [CrossRef]
77. Needham, B.L.; Rehkopf, D.; Adler, N.; Gregorich, S.; Lin, J.; Blackburn, E.H.; Epel, E. Leukocyte telomere length and mortality in the national health and nutrition examination survey, 1999-2002. *Epidemiology* **2015**, *26*, 528–535. [CrossRef] [PubMed]
78. Deelen, J.; Beekman, M.; Codd, V.; Trompet, S.; Broer, L.; Hagg, S.; Fischer, K.; Thijssen, P.E.; Suchiman, H.E.D.; Postmus, I.; et al. Leukocyte telomere length associates with prospective mortality independent of immune-related parameters and known genetic markers. *Int. J. Epidemiol.* **2014**, *43*, 878–886. [CrossRef]
79. Fulop, T.; Larbi, A.; Dupuis, G.; Le Page, A.; Frost, E.H.; Cohen, A.A.; Witkowski, J.; Franceschi, C. Immunosenescence and inflamm-aging as two sides of the same coin: Friends or Foes? *Front. Immunol.* **2018**, *8*, 1960. [CrossRef]
80. van der Harst, P.; van der Steege, G.; de Boer, R.A.; Voors, A.A.; Hall, A.S.; Mulder, M.J.; van Gilst, W.; van Veldhuisen, D. Telomere length of circulating leukocytes is decreased in patients with chronic heart failure. *J. Am. Coll. Cardiol.* **2007**, *49*, 1459–1464. [CrossRef]
81. Forero, D.A.; Gonzalez-Giraldo, Y.; Lopez-Quintero, C.; Castro-Vega, L.J.; Barreto, G.E.; Perry, G. Meta-analysis of telomere length in Alzheimer's disease. *J. Gerontol. Ser. A Biol. Sci. Med Sci.* **2016**, *71*, 1069–1073. [CrossRef]
82. Forero, D.A.; Gonzalez-Giraldo, Y.; Lopez-Quintero, C.; Castro-Vega, L.J.; Barreto, G.E.; Perry, G. Telomere length in Parkinson's disease: A meta-analysis. *Exp. Gerontol.* **2016**, *75*, 53–55. [CrossRef] [PubMed]
83. Panossian, L.A.; Porter, V.R.; Valenzuela, H.F.; Zhu, X.; Reback, E.; Masterman, D.; Cummings, J.L.; Effros, R.B. Telomere shortening in T cells correlates with Alzheimer's disease status. *Neurobiol. Aging* **2003**, *24*, 77–84. [CrossRef]
84. D'Mello, M.J.J.; Ross, S.A.; Briel, M.; Anand, S.S.; Gerstein, H.; Pare, G. Association between shortened leukocyte telomere length and cardiometabolic outcomes: Systematic review and meta-analysis. *Circ. Cardiovasc. Genet.* **2015**, *8*, 82–90. [CrossRef]
85. Dai, D.F.; Chiao, Y.A.; Marcinek, D.J.; Szeto, H.H.; Rabinovitch, P.S. Mitochondrial oxidative stress in aging and healthspan. *Longev. Healthspan* **2014**, *3*, 6. [CrossRef] [PubMed]
86. Serra, V.; von Zglinicki, T.; Lorenz, M.; Saretzki, G. Extracellular superoxide dismutase is a major antioxidant in human fibroblasts and slows telomere shortening. *J. Biol. Chem.* **2003**, *278*, 6824–6830. [CrossRef] [PubMed]
87. Liu, L.; Trimarchi, J.R.; Smith, P.J.S.; Keefe, D.L. Mitochondrial dysfunction leads to telomere attrition and genomic instability. *Aging Cell* **2002**, *1*, 40–46. [CrossRef]
88. Shay, J.W. Role of telomeres and telomerase in aging and cancer. *Cancer Discov.* **2016**, *6*, 584–593. [CrossRef]
89. Savage, S.A.; Gadalla, S.M.; Chanock, S.J. The long and short of telomeres and cancer association studies. *JNCI J. Natl. Cancer Inst.* **2013**, *105*, 448–449. [CrossRef] [PubMed]
90. Mensa, E.; Latini, S.; Ramini, D.; Storci, G.; Bonafe, M.; Olivieri, F. The telomere world and aging: Analytical challenges and future perspectives. *Ageing Res. Rev.* **2019**, *50*, 27–42. [CrossRef]
91. He, J.; Tu, C.; Liu, Y. Role of lncRNAs in aging and age-related diseases. *Aging Med.* **2018**, *1*, 158–175. [CrossRef]
92. Huan, T.; Chen, G.; Liu, C.; Bhattacharya, A.; Rong, J.; Chen, B.H.; Seshadri, S.; Tanriverdi, K.; Freedman, J.E.; Larson, M.G.; et al. Age-associated microRNA expression in human peripheral blood is associated with all-cause mortality and age-related traits. *Aging Cell* **2018**, *17*, e12687. [CrossRef]
93. Gupta, S.C.; Tripathi, Y.N. Potential of long non-coding RNAs in cancer patients: From biomarkers to therapeutic targets. *Int. J. Cancer* **2017**, *140*, 1955–1967. [CrossRef] [PubMed]
94. Abdelmohsen, K.; Panda, A.; Kang, M.J.; Xu, J.; Selimyan, R.; Yoon, J.H.; Martindale, J.L.; De, S.; Wood, W.H.; Becker, K.G.; et al. Senescence-associated lncRNAs: Senescence-associated long noncoding RNAs. *Aging Cell* **2013**, *12*, 890–900. [CrossRef] [PubMed]
95. Frenk, S.; Houseley, J. Gene expression hallmarks of cellular ageing. *Biogerontology* **2018**, *19*, 547–566. [CrossRef]
96. Westerterp, M.; Tsuchiya, K.; Tattersall, I.W.; Fotakis, P.; Bochem, A.E.; Molusky, M.M.; Ntonga, V.; Abramowicz, S.; Parks, J.S.; Welch, C.L.; et al. Deficiency of ATP-binding cassette transporters A1 and G1 in endothelial cells accelerates atherosclerosis in mice. *Arterioscler. Thromb. Vasc. Biol.* **2016**, *36*, 1328–1337. [CrossRef]
97. D'Aquila, P.; Crocco, P.; De Rango, F.; Indiveri, C.; Bellizzi, D.; Rose, G.; Passarino, G. A genetic variant of ASCT2 hampers in vitro RNA splicing and correlates with human longevity. *Rejuvenation Res.* **2018**, *21*, 193–199. [CrossRef] [PubMed]
98. Klucken, J.; Buchler, C.; Orso, E.; Kaminski, W.E.; Porsch-Ozcurumez, M.; Liebisch, C.; Kapinsky, M.; Diederich, W.; Drobnik, W.; Dean, M.; et al. ABCG1 (ABC8), the human homolog of the Drosophila white gene, is a regulator of macrophage cholesterol and phospholipid transport. *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 817–822. [CrossRef] [PubMed]
99. Balliu, B.; Durrant, M.; de Goede, O.; Abell, N.; Li, X.; Liu, B. Genetic dysregulation of gene expression and splicing during a ten-year period of human aging. *bioRxiv* **2019**, 519520. [CrossRef]
100. Nakamura, S.; Kawai, K.; Takeshita, Y.; Honda, M.; Takamura, T.; Kaneko, S.; Matoba, R.; Matsubara, K. Identification of blood biomarkers of aging by transcript profiling of whole blood. *Biochem. Biophys. Res. Commun.* **2012**, *418*, 313–318. [CrossRef]
101. Harries, L.W.; Hernandez, D.; Henley, W.; Wood, A.R.; Holly, A.C.; Bradley-Smith, R.M.; Yaghootkar, H.; Dutta, A.; Murray, A.; Frayling, T.M.; et al. Human aging is characterized by focused changes in gene expression and deregulation of alternative splicing. *Aging Cell* **2011**, *10*, 868–878. [CrossRef] [PubMed]
102. Holly, A.C.; Melzer, D.; Pilling, L.C.; Henley, W.; Hernandez, D.G.; Singleton, A.B.; Bandinelli, S.; Guralnik, J.M.; Ferrucci, L.; Harries, L.W. Towards a gene expression biomarker set for human biological age. *Aging Cell* **2013**, *12*, 324–326. [CrossRef] [PubMed]
103. Dhahbi, J.M. Circulating small noncoding RNAs as biomarkers of aging. *Ageing Res. Rev.* **2014**, *17*, 86–98. [CrossRef] [PubMed]

104. Dumortier, O.; Hinault, C.; Van Obberghen, E. MicroRNAs and metabolism crosstalk in energy homeostasis. *Cell Metab.* **2013**, *18*, 312–324. [CrossRef]

105. Hayes, J.; Peruzzi, P.P.; Lawler, S. MicroRNAs in cancer: Biomarkers, functions and therapy. *Trends Mol. Med.* **2014**, *20*, 460–469. [CrossRef]

106. Jung, M.; Pfeifer, G.P. Aging and DNA methylation. *BMC Biol.* **2015**, *13*, 7. [CrossRef]

107. Huan, T.; Rong, J.; Tanriverdi, K.; Meng, Q.; Bhattacharya, A.; McManus, D.D.; Joehanes, R.; Assimes, T.L.; McPherson, R.; Samani, N.J.; et al. Dissecting the roles of microRNAs in coronary heart disease via integrative genomic analyses. *Arterioscler. Thromb. Vasc. Biol.* **2015**, *35*, 1011–1021. [CrossRef] [PubMed]

108. Huan, T.; Rong, J.; Liu, C.; Zhang, X.; Tanriverdi, K.; Joehanes, R.; Chen, B.H.; Murabito, J.M.; Yao, C.; Courchesne, P.; et al. Genome-wide identification of microRNA expression quantitative trait loci. *Nat. Commun.* **2015**, *6*, 7601. [CrossRef]

109. Shi, L.; Liao, J.; Liu, B.; Zeng, F.; Zhang, L. Mechanisms and therapeutic potential of microRNAs in hypertension. *Drug Discov. Today* **2015**, *20*, 1188–1204. [CrossRef]

110. Iacomino, G.; Siani, A. Role of microRNAs in obesity and obesity-related diseases. *Genes Nutr.* **2017**, *12*, 23. [CrossRef]

111. Feng, J.; Xing, W.; Xie, L. Regulatory roles of microRNAs in diabetes. *Int. J. Mol. Sci.* **2016**, *17*, 1729. [CrossRef] [PubMed]

112. Noren Hooten, N.; Fitzpatrick, M.; Wood, W.H., III; De, S.; Ejiogu, N.; Zhang, Y.; Mattison, J.A.; Becker, K.G.; Zonderman, A.B.; Evans, M.K.; et al. Age-related changes in microRNA levels in serum. *Aging* **2013**, *5*, 725–740. [CrossRef] [PubMed]

113. ElSharawy, A.; Keller, A.; Flachsbart, F.; Wendschlag, A.; Jacobs, G.; Kefer, N.; Brefort, T.; Leidinger, P.; Backes, C.; Meese, E.; et al. Genome-wide miRNA signatures of human longevity. *Aging Cell* **2012**, *11*, 607–616. [CrossRef] [PubMed]

114. Noren Hooten, N.; Abdelmohsen, K.; Gorospe, M.; Ejiogu, N.; Zonderman, A.B.; Evans, M.K. MicroRNA expression patterns reveal differential expression of target genes with age. *PLoS ONE* **2010**, *5*, e10724. [CrossRef]

115. Kinser, H.E.; Pincus, Z. MicroRNAs as modulators of longevity and the aging process. *Hum. Genet.* **2020**, *139*, 291–308. [CrossRef]

116. Eyileten, C.; Wicik, Z.; De Rosa, S.; Mirowska-Guzel, D.; Soplinska, A.; Indolfi, C.; Jastrzebska-Kurkowska, I.; Czlonkowska, A.; Postula, M. MicroRNAs as diagnostic and prognostic biomarkers in ischemic stroke-A comprehensive review and bioinformatic analysis. *Cells* **2018**, *7*, 249. [CrossRef]

117. Roy-O'Reilly, M.A.; Ahnstedt, H.; Spychala, M.S.; Munshi, Y.; Aronowski, J.; Sansing, L.H.; McCullough, L.D. Aging exacerbates neutrophil pathogenicity in ischemic stroke. *Aging* **2020**, *12*, 436–461. [CrossRef]

118. Li, X.; Khanna, A.; Li, N.; Wang, E. Circulatory miR-34a as an RNA-based, noninvasive biomarker for brain aging. *Aging* **2011**, *3*, 985–1002. [CrossRef] [PubMed]

119. Olivieri, F.; Spazzafumo, L.; Santini, G.; Lazzarini, R.; Albertini, M.C.; Rippo, M.R.; Galeazzi, R.; Abbatecola, A.M.; Marcheselli, F.; Monti, D.; et al. Age-related differences in the expression of circulating microRNAs: miR-21 as a new circulating marker of inflammaging. *Mech. Ageing Dev.* **2012**, *133*, 675–685. [CrossRef]

120. Kumar, S.; Vijayan, M.; Reddy, P.H. MicroRNA-455-3p as a potential peripheral biomarker for Alzheimer's disease. *Hum. Mol. Genet.* **2017**, *26*, 3808–3822. [CrossRef]

121. Kumar, S.; Reddy, A.P.; Yin, X.; Reddy, P.H. Novel MicroRNA-455-3p and its protective effects against abnormal APP processing and amyloid beta toxicity in Alzheimer's disease. *Biochim. Biophys. Acta Mol. Basis Dis.* **2019**, *1865*, 2428–2440. [CrossRef] [PubMed]

122. Fatica, A.; Bozzoni, I. Long non-coding RNAs: New players in cell differentiation and development. *Nat. Rev. Genet.* **2014**, *15*, 7–21. [CrossRef]

123. Jin, L.; Song, Q.; Zhang, W.; Geng, B.; Cai, J. Roles of long noncoding RNAs in aging and aging complications. *Biochim. Biophys Acta Mol. Basis Dis.* **2019**, *1865*, 1763–1771. [CrossRef]

124. Pereira Fernandes, D.; Bitar, M.; Jacobs, F.M.J.; Barry, G. Long non-coding RNAs in neuronal aging. *Non-Coding RNA* **2018**, *4*, 12. [CrossRef]

125. Boon, R.A.; Hofmann, P.; Michalik, K.M.; Lozano-Vidal, N.; Berghaeuser, D.; Fischer, A.; Knau, A.; Jae, N.; Schuermann, C.; Dimmeler, S. Long noncoding RNA Meg3 controls endothelial cell aging and function implications for regenerative angiogenesis. *J. Am. Coll. Cardiol.* **2016**, *68*, 2589–2591. [CrossRef] [PubMed]

126. Knupp, D.; Miura, P. CircRNA accumulation: A new hallmark of aging? *Mech. Ageing Dev.* **2018**, *173*, 71–79. [CrossRef]

127. Alhasan, A.A.; Izuogu, O.G.; Al-Balool, H.H.; Steyn, J.S.; Evans, A.; Colzani, M.; Ghevaert, C.; Mountford, J.C.; Marenah, L.; Elliott, D.J.; et al. Circular RNA enrichment in platelets is a signature of transcriptome degradation. *Blood* **2016**, *127*, E1–E11. [CrossRef]

128. Memczak, S.; Papavasileiou, P.; Peters, O.; Rajewsky, N. Identification and characterization of circular RNAs as a new class of putative biomarkers in human blood. *PLoS ONE* **2015**, *10*, e141214. [CrossRef]

129. Koh, W.; Pan, W.; Gawad, C.; Fan, H.C.; Kerchner, G.A.; Wyss-Coray, T.; Blumenfeld, Y.J.; El-Sayed, Y.Y.; Quake, S.R. Noninvasive in vivo monitoring of tissue-specific global gene expression in humans. *Proc. Natl. Acad. Sci. USA* **2014**, *111*, 7361–7366. [CrossRef] [PubMed]

130. Bahn, J.H.; Zhang, Q.; Li, F.; Chan, T.M.; Lin, X.; Kim, Y.; Wong, D.T.W.; Xiao, X. The landscape of microRNA, piwi-interacting RNA, and circular RNA in human saliva. *Clin. Chem.* **2015**, *61*, 221–230. [CrossRef]

131. Pan, T.; Sun, X.Q.; Liu, Y.X.; Li, H.; Deng, G.B.; Lin, H.H.; Wang, S. Heat stress alters genome-wide profiles of circular RNAs in Arabidopsis. *Plant Mol. Biol.* **2018**, *96*, 217–229. [CrossRef] [PubMed]

132. Rybak-Wolf, A.; Stottmeister, C.; Glazar, P.; Jens, M.; Pino, N.; Giusti, S.; Hanan, M.; Behm, M.; Bartok, O.; Ashwal-Fluss, R.; et al. Circular RNAs in the mammalian brain are highly abundant, conserved, and dynamically expressed. *Mol. Cell* **2015**, *58*, 870–885. [CrossRef]

133. Fanciulli, A.; Wenning, G.K. Multiple-system atrophy. *N. Engl. J. Med.* **2015**, *372*, 249–263. [CrossRef]

134. Pastushkova, L.K.; Kononikhin, A.S.; Tiys, E.S.; Dobrokhotov, I.V.; Ivanisenko, V.A.; Nikolaev, E.N.; Larina, I.M.; Popov, I.A. Characteristics of age-dependent changes in urine proteome in healthy men. *Adv. Gerontol.* **2016**, *6*, 123–128. [CrossRef]

135. Diz, A.P.; Martinez-Fernandez, M.; Rolan-Alvarez, E. Proteomics in evolutionary ecology: Linking the genotype with the phenotype. *Mol. Ecol.* **2012**, *21*, 1060–1080. [CrossRef]

136. Semba, R.D.; Zhang, P.; Zhu, M.; Fabbri, E.; Gonzalez-Freire, M.; Moaddel, R.; Geng-Spyropoulos, M.; Ferrucci, L. A targeted proteomic assay for the measurement of plasma proteoforms related to human aging phenotypes. *Proteomics* **2017**, *17*, 1600232. [CrossRef] [PubMed]

137. Mehra, V.C.; Ramgolam, V.S.; Bender, J.R. Cytokines and cardiovascular disease. *J. Leukoc. Biol.* **2005**, *78*, 805–818. [CrossRef]

138. Swardfager, W.; Lanctot, K.; Rothenburg, L.; Wong, A.; Cappell, J.; Herrmann, N. A meta-analysis of cytokines in Alzheimer's disease. *Biol. Psychiatry* **2010**, *68*, 930–941. [CrossRef]

139. Menni, C.; Kiddle, S.J.; Mangino, M.; Vinuela, A.; Psatha, M.; Steves, C.; Sattlecker, M.; Buil, A.; Newhouse, S.; Nelson, S.; et al. Circulating proteomic signatures of chronological age. *J. Gerontol. Ser. A Biol. Sci. Med. Sci.* **2015**, *70*, 809–816. [CrossRef] [PubMed]

140. Santos-Lozano, A.; Valenzuela, P.L.; Llavero, F.; Lista, S.; Carrera-Bastos, P.; Hampel, H.; Pareja-Galeano, H.; Galvez, B.G.; Lopez, J.A.; Vazquez, J.; et al. Successful aging: Insights from proteome analyses of healthy centenarians. *Aging* **2020**, *12*, 3502–3515. [CrossRef] [PubMed]

141. Sebastiani, P.; Federico, A.; Morris, M.; Gurinovich, A.; Tanaka, T.; Chandler, K.B.; Andersen, S.L.; Denis, G.; Costello, K.; Ferrucci, L.; et al. Protein signatures of centenarians and their offspring suggest centenarians age slower than other humans. *Aging Cell* **2021**, *20*, e13290. [CrossRef] [PubMed]

142. Basisty, N.; Kale, A.; Jeon, O.H.; Kuehnemann, C.; Payne, T.; Rao, C.; Holtz, A.; Shah, S.; Sharma, V.; Ferrucci, L.; et al. A proteomic atlas of senescence-associated secretomes for aging biomarker development. *PLoS Biol.* **2020**, *18*, e3000599. [CrossRef] [PubMed]

143. Conte, M.; Martucci, M.; Chiariello, A.; Franceschi, C.; Salvioli, S. Mitochondria, immunosenescence and inflammaging: A role for mitokines? *Semin. Immunopathol.* **2020**, *42*, 607–617. [CrossRef] [PubMed]

144. Coppe, J.P.; Patil, C.K.; Rodier, F.; Sun, Y.; Munoz, D.P.; Goldstein, J.; Nelson, P.S.; Desprez, P.Y.; Campisi, J. Senescence-associated secretory phenotypes reveal cell-nonautonomous functions of oncogenic RAS and the p53 tumor suppressor. *PLoS Biol.* **2008**, *6*, 2853–2868. [CrossRef] [PubMed]

145. Campisi, J. Cellular senescence: Putting the paradoxes in perspective. *Curr. Opin. Genet. Dev.* **2011**, *21*, 107–112. [CrossRef]

146. Rodier, F.; Coppe, J.P.; Patil, C.K.; Hoeijmakers, W.A.M.; Munoz, D.P.; Raza, S.R.; Freund, A.; Campeau, E.; Davalos, A.; Campisi, J. Persistent DNA damage signalling triggers senescence-associated inflammatory cytokine secretion. *Nat. Cell Biol.* **2009**, *11*, 973–979. [CrossRef] [PubMed]

147. Orjalo, A.V.; Bhaumik, D.; Gengler, B.K.; Scott, G.K.; Campisi, J. Cell surface-bound IL-1 alpha is an upstream regulator of the senescence-associated IL-6/IL-8 cytokine network. *Proc. Natl. Acad. Sci. USA* **2009**, *106*, 17031–17036. [CrossRef] [PubMed]

148. Freund, A.; Patil, C.K.; Campisi, J. p38MAPK is a novel DNA damage response-independent regulator of the senescence-associated secretory phenotype. *Embo J.* **2011**, *30*, 1536–1548. [CrossRef]

149. Laberge, R.M.; Sun, Y.; Orjalo, A.V.; Patil, C.K.; Freund, A.; Zhou, L.; Curran, S.C.; Davalos, A.R.; Wilson-Edell, K.A.; Liu, S.; et al. MTOR regulates the pro-tumorigenic senescence-associated secretory phenotype by promoting IL1A translation. *Nat. Cell Biol.* **2015**, *1548*, 1049–1061. [CrossRef] [PubMed]

150. Watroba, M.; Szukiewicz, D. The role of sirtuins in aging and age-related diseases. *Adv. Med. Sci.* **2016**, *61*, 52–62. [CrossRef] [PubMed]

151. Watroba, M.; Dudek, I.; Skoda, M.; Stangret, A.; Rzodkiewicz, P.; Szukiewicz, D. Sirtuins, epigenetics and longevity. *Ageing Res. Rev.* **2017**, *40*, 11–19. [CrossRef] [PubMed]

152. Salminen, A.; Kaarniranta, K. AMP-activated protein kinase (AMPK) controls the aging process via an integrated signaling network. *Ageing Res. Rev.* **2012**, *11*, 230–241. [CrossRef] [PubMed]

153. Kenyon, C.J. The genetics of ageing. *Nature* **2010**, *464*, 504–512. [CrossRef]

154. Dang, W.; Steffen, K.K.; Perry, R.; Dorsey, J.A.; Johnson, F.B.; Shilatifard, A.; Kaeberlein, M.; Kennedy, B.K.; Berger, S.L. Histone H4 lysine 16 acetylation regulates cellular lifespan. *Nature* **2009**, *459*, 802–807. [CrossRef]

155. Blackburn, E.H. Switching and signaling at the telomere. *Cell* **2001**, *106*, 661–673. [CrossRef]

156. Jazwinski, S.M.; Yashin, A.I. Aging and health—A systems biology perspective. Introduction. *Interdiscip. Top. Gerontol.* **2015**, *40*, 7–12. [CrossRef]

157. Bjedov, I.; Partridge, L. A longer and healthier life with TOR down-regulation: Genetics and drugs. *Biochem. Soc. Trans.* **2011**, *39*, 460–465. [CrossRef] [PubMed]

158. Vanhooren, V.; Santos, A.N.; Voutetakis, K.; Petropoulos, I.; Libert, C.; Simm, A.; Gonos, E.S.; Friguet, B. Protein modification and maintenance systems as biomarkers of ageing. *Mech. Ageing Dev.* **2015**, *151*, 71–84. [CrossRef]

159. Oien, D.B.; Moskovitz, J. Substrates of the methionine sulfoxide reductase system and their physiological relevance. *Curr. Top. Dev. Biol.* **2008**, *80*, 93–133. [CrossRef] [PubMed]

160. Srivastava, S. Emerging insights into the metabolic alterations in aging using metabolomics. *Metabolites* **2019**, *9*, 301. [CrossRef]
161. Wang, Z.; Bian, L.; Mo, C.; Shen, H.; Zhao, L.J.; Su, K.J.; Kukula, M.; Lee, J.T.; Armstrong, D.W.; Recker, R.; et al. Quantification of aminobutyric acids and their clinical applications as biomarkers for osteoporosis. *Commun. Biol.* **2020**, *3*, 39. [CrossRef]
162. Jylhava, J.; Pedersen, N.L.; Hagg, S. Biological age predictors. *EBioMedicine* **2017**, *21*, 29–36. [CrossRef]
163. Yaneske, E.; Angione, C. The poly-omics of ageing through individual-based metabolic modelling. *BMC Bioinform.* **2018**, *19*, 415–496. [CrossRef]
164. Rivero-Segura, N.A.; Bello-Chavolla, O.Y.; Barrera-Vazquez, O.S.; Gutierrez-Robledo, L.M.; Gomez-Verjan, J.C. Promising biomarkers of human aging: In search of a multi-omics panel to understand the aging process from a multidimensional perspective. *Ageing Res. Rev.* **2020**, *64*, 101164. [CrossRef] [PubMed]
165. Zierer, J.; Menni, C.; Kastenmuler, G.; Spector, T.D. Integration of 'omics' data in aging research: From biomarkers to systems biology. *Aging Cell* **2015**, *14*, 933–944. [CrossRef]
166. Robinson, O.; Chadeau Hyam, M.; Karaman, I.; Climaco Pinto, R.; Ala-Korpela, M.; Handakas, E.; Fiorito, G.; Gao, H.; Heard, A.; Jarvelin, M.R.; et al. Determinants of accelerated metabolomic and epigenetic aging in a UK cohort. *Aging Cell* **2020**, *19*, 19. [CrossRef] [PubMed]
167. Jove, M.; Mate, I.; Naudi, A.; Mota-Martorell, N.; Portero-Otin, M.; De la Fuente, M.; Pamplona, R. Human aging is a metabolome-related matter of gender. *J. Gerontol. Ser. A Biol. Sci. Med. Sci.* **2016**, *71*, 578–585. [CrossRef] [PubMed]
168. Kenyon, C. The plasticity of aging: Insights from long-lived mutants. *Cell* **2005**, *120*, 449–460. [CrossRef]
169. Fontana, L.; Partridge, L.; Longo, V.D. Extending Healthy Life Span-From Yeast to Humans. *Science* **2010**, *328*, 321–326. [CrossRef] [PubMed]
170. Junnila, R.K.; List, E.O.; Berryman, D.E.; Murrey, J.W.; Kopchick, J.J. The GH/IGF-1 axis in ageing and longevity. *Nat. Rev. Endocrinol.* **2013**, *9*, 366–376. [CrossRef]
171. Massudi, H.; Grant, R.; Braidy, N.; Guest, J.; Farnsworth, B.; Guillemin, G.J. Age-associated changes In oxidative stress and NAD(+) metabolism in human tissue. *PLoS ONE* **2012**, *7*, e42357. [CrossRef]
172. Verdin, E. NAD(+) in aging, metabolism, and neurodegeneration. *Science* **2015**, *350*, 1208–1213. [CrossRef]
173. Mendelsohn, A.R.; Larrick, J.W. The NAD+/PARP1/SIRT1 axis in aging. *Rejuvenation Res.* **2017**, *20*, 244–247. [CrossRef] [PubMed]
174. Zhang, H.; Ryu, D.; Wu, Y.; Gariani, K.; Wang, X.; Luan, P.; D'Amico, D.; Ropelle, E.R.; Lutolf, M.P.; Aebersold, R.; et al. NAD(+) repletion improves mitochondrial and stem cell function and enhances life span in mice. *Science* **2016**, *352*, 1436–1443. [CrossRef]
175. Igarashi, M.; Miura, M.; Williams, E.; Jaksch, F.; Kadowaki, T.; Yamauchi, T.; Guarente, L. NAD(+) supplementation rejuvenates aged gut adult stem cells. *Aging Cell* **2019**, *18*, e12935. [CrossRef]
176. Nacarelli, T.; Lau, L.; Fukumoto, T.; Zundell, J.; Fatkhutdinov, N.; Wu, S.; Aird, K.M.; Iwasaki, O.; Kossenkov, A.V.; Schultz, D.; et al. NAD(+) metabolism governs the proinflammatory senescence-associated secretome. *Nat. Cell Biol.* **2019**, *21*, 397–407. [CrossRef] [PubMed]
177. Baker, D.J.; Wijshake, T.; Tchkonia, T.; LeBrasseur, N.K.; Childs, B.G.; van de Sluis, B.; Kirkland, J.L.; van Deursen, J.M. Clearance of p16(Ink4a)-positive senescent cells delays ageing-associated disorders. *Nature* **2011**, *479*, 232–236. [CrossRef] [PubMed]
178. Zhang, G.; Li, J.; Purkayastha, S.; Tang, Y.; Zhang, H.; Yin, Y.; Li, B.; Liu, G.; Cai, D.S. Hypothalamic programming of systemic ageing involving IKK-beta, NF-kappa B and GnRH. *Nature* **2013**, *497*, 211–216. [CrossRef]
179. Melendez, A.; Talloczy, Z.; Seaman, M.; Eskelinen, E.L.; Hall, D.H.; Levine, B. Autophagy genes are essential for dauer development and life-span extension in C-elegans. *Science* **2003**, *301*, 1387–1391. [CrossRef] [PubMed]
180. Simonsen, A.; Cumming, R.C.; Brech, A.; Isakson, P.; Schubert, D.R.; Finley, K.D. Promoting basal levels of autophagy in the nervous system enhances longevity and oxidant resistance in adult Drosophila. *Autophagy* **2008**, *4*, 176–184. [CrossRef]
181. Pyo, J.O.; Yoo, S.M.; Ahn, H.H.; Nah, J.; Hong, S.H.; Kam, T.I.; Jung, S.; Jung, Y.K. Overexpression of Atg5 in mice activates autophagy and extends lifespan. *Nat. Commun.* **2013**, *4*, 2300. [CrossRef]
182. Almajwal, A.M.; Alam, I.; Abulmeaty, M.; Razak, S.; Pawelec, G.; Alam, W. Intake of dietary advanced glycation end products influences inflammatory markers, immune phenotypes, and antiradical capacity of healthy elderly in a little-studied population. *Food Sci. Nutr.* **2020**, *8*, 1046–1057. [CrossRef]
183. Grosskopf, A.; Simm, A. Carbohydrates in nutrition: Friend or foe? *Z. Gerontol. Geriatr.* **2020**, *53*, 290–294. [CrossRef]
184. Chak, C.M.; Lacruz, M.E.; Adam, J.; Brandmaier, S.; Covic, M.; Huang, J.; Meisinger, C.; Tiller, D.; Prehn, C.; Adamski, J.; et al. Ageing investigation using two-time-point metabolomics data from KORA and CARLA studies. *Metabolites* **2019**, *9*, 44. [CrossRef] [PubMed]
185. Deelen, J.; Kettunen, J.; Fischer, K.; van der Spek, A.; Trompet, S.; Kastenmueller, G.; Boyd, A.; Zierer, J.; van den Akker, E.B.; Ala-Korpela, M.; et al. A metabolic profile of all-cause mortality risk identified in an observational study of 44,168 individuals. *Nat. Commun.* **2019**, *10*, 3346. [CrossRef] [PubMed]
186. Kohler, I.; Verhoeven, A.; Derks, R.J.E.; Giera, M. Analytical pitfalls and challenges in clinical metabolomics. *Bioanalysis* **2016**, *8*, 1509–1532. [CrossRef] [PubMed]
187. Sanchez-Morate, E.; Gimeno-Mallench, L.; Stromsnes, K.; Sanz-Ros, J.; Roman-Dominguez, A.; Parejo-Pedrajas, S.; Ingles, M.; Olaso, G.; Gambini, J.; Mas-Bargues, C. Relationship between diet, microbiota, and healthy aging. *Biomedicines* **2020**, *8*, 287. [CrossRef]
188. Salvador Barrera-Vazquez, O.; Carlos Gomez-Verjan, J. The unexplored world of human virome, mycobiome, and archaeome in aging. *J. Gerontol. Ser. A Biol. Sci. Med. Sci.* **2020**, *75*, 1834–1837. [CrossRef]

189. Aleman, F.D.D.; Valenzano, D.R. Microbiome evolution during host aging. *PLoS Pathog.* **2019**, *15*, e1007727. [CrossRef]
190. Finlay, B.B.; Pettersson, S.; Melby, M.K.; Bosch, T.C.G. The microbiome mediates environmental effects on aging. *BioEssays* **2019**, *41*, e1800257. [CrossRef]
191. Lynch, S.V.; Pedersen, O. The human intestinal microbiome in health and disease. *N. Engl. J. Med.* **2016**, *375*, 2369–2379. [CrossRef]
192. Badal, V.D.; Vaccariello, E.D.; Murray, E.R.; Yu, K.E.; Knight, R.; Jeste, D.V.; Nguyen, T.T. The gut microbiome, aging, and longevity: A systematic review. *Nutrients* **2020**, *12*, 3759. [CrossRef] [PubMed]
193. Claesson, M.J.; Cusack, S.; O'Sullivan, O.; Greene-Diniz, R.; de Weerd, H.; Flannery, E.; Marchesi, J.; Falush, D.; Dinan, T.; Fitzgerald, G.; et al. Composition, variability, and temporal stability of the intestinal microbiota of the elderly. *Proc. Natl. Acad. Sci. USA* **2011**, *108*, 4586–4591. [CrossRef]
194. Thomas, V.; Clark, J.; Dore, J. Fecal microbiota analysis: An overview of sample collection methods and sequencing strategies. *Future Microbiol.* **2015**, *10*, 1485–1504. [CrossRef]
195. Kumar, M.; Babaei, P.; Ji, B.; Nielsen, J. Human gut microbiota and healthy aging: Recent developments and future prospective. *Nutr. Healthy Aging* **2016**, *4*, 3–16. [CrossRef]
196. Odamaki, T.; Kato, K.; Sugahara, H.; Hashikura, N.; Takahashi, S.; Xiao, J.Z.; Abe, F.; Osawa, R. Age-related changes in gut microbiota composition from newborn to centenarian: A cross-sectional study. *BMC Microbiol.* **2016**, *16*, 90. [CrossRef]
197. Hopkins, M.J.; Sharp, R.; Macfarlane, G.T. Variation in human intestinal microbiota with age. *Dig. Liver Dis.* **2002**, *34*, S12–S18. [CrossRef]
198. Vaiserman, A.M.; Koliada, A.K.; Marotta, F. Gut microbiota: A player in aging and a target for anti-aging intervention. *Ageing Res. Rev.* **2017**, *35*, 36–45. [CrossRef]
199. O'Hagan, C.; Li, J.V.; Marchesi, J.R.; Plummer, S.; Garaiova, I.; Good, M.A. Long-term multi-species Lactobacillus and Bifidobacterium dietary supplement enhances memory and changes regional brain metabolites in middle-aged rats. *Neurobiol. Learn. Mem.* **2017**, *144*, 36–47. [CrossRef] [PubMed]
200. Rondanelli, M.; Giacosa, A.; Faliva, M.A.; Perna, S.; Allieri, F.; Castellazzi, A.M. Review on microbiota and effectiveness of probiotics use in older. *World J. Clin. Cases* **2015**, *3*, 156–162. [CrossRef]
201. Han, B.; Sivaramakrishnan, P.; Lin, C.C.J.; Neve, I.A.A.; He, J.; Tay, L.W.R.; Sowa, J.N.; Sizovs, A.; Du, G.W.; Wang, J.; et al. Microbial genetic composition tunes host longevity. *Cell* **2017**, *169*, 1249–1262. [CrossRef] [PubMed]
202. An, R.; Wilms, E.; Masclee, A.A.M.; Smidt, H.; Zoetendal, E.G.; Jonkers, D. Age-dependent changes in GI physiology and microbiota: Time to reconsider? *Gut* **2018**, *67*, 2213–2222. [CrossRef] [PubMed]
203. Wu, G.D.; Chen, J.; Hoffmann, C.; Bittinger, K.; Chen, Y.Y.; Keilbaugh, S.A.; Bewtra, M.; Knights, D.; Walters, W.A.; Knight, R.; et al. Linking long-term dietary patterns with gut microbial enterotypes. *Science* **2011**, *334*, 105–108. [CrossRef]
204. Yatsunenko, T.; Rey, F.E.; Manary, M.J.; Trehan, I.; Dominguez-Bello, M.G.; Contreras, M.; Magris, M.; Hidalgo, G.; Baldassano, R.N.; Anokhin, A.P.; et al. Human gut microbiome viewed across age and geography. *Nature* **2012**, *486*, 222–227. [CrossRef]
205. Wu, G.D.; Lewis, J.D. Analysis of the human gut microbiome and association with disease. *Clin. Gastroenterol. Hepatol.* **2013**, *11*, 774–777. [CrossRef] [PubMed]
206. Musso, G.; Gambino, R.; Cassader, M. Interactions between gut microbiota and host metabolism predisposing to obesity and diabetes. *Annu. Rev. Med.* **2011**, *62*, 361–380. [CrossRef] [PubMed]
207. Schneiderhan, J.; Master-Hunter, T.; Locke, A. Targeting gut flora to treat and prevent disease. *J. Fam. Pract.* **2016**, *65*, 33–38.
208. Rodriguez-Castano, G.P.; Caro-Quintero, A.; Reyes, A.; Lizcano, F. Advances in gut microbiome research, opening new strategies to cope with a western lifestyle. *Front. Genet.* **2017**, *7*, 224. [CrossRef] [PubMed]
209. Candela, M.; Biagi, E.; Brigidi, P.; O'Toole, P.W.; De Vos, W.M. Maintenance of a healthy trajectory of the intestinal microbiome during aging: A dietary approach. *Mech. Ageing Dev.* **2014**, *136*, 70–75. [CrossRef]
210. Marchesi, J.R.; Adams, D.H.; Fava, F.; Hermes, G.D.A.; Hirschfield, G.M.; Hold, G.; Quraishi, M.N.; Kinross, J.; Smidt, H.; Tuohy, K.M.; et al. The gut microbiota and host health: A new clinical frontier. *Gut* **2016**, *65*, 330–339. [CrossRef]
211. Buford, T.W. (Dis)Trust your gut: The gut microbiome in age-related inflammation, health, and disease. *Microbiome* **2017**, *5*, 80. [CrossRef]
212. Franceschi, C.; Garagnani, P.; Parini, P.; Giuliani, C.; Santoro, A. Inflammaging: A new immune-metabolic viewpoint for age-related diseases. *Nat. Rev. Endocrinol.* **2018**, *14*, 576–590. [CrossRef]
213. Thevaranjan, N.; Puchta, A.; Schulz, C.; Naidoo, A.; Szamosi, J.C.; Verschoor, C.P.; Loukov, D.; Schenck, L.P.; Jury, J.; Foley, K.; et al. Age-associated microbial dysbiosis promotes intestinal permeability, systemic inflammation, and macrophage dysfunction. *Cell Host Microbe* **2017**, *21*, 455–466. [CrossRef]
214. Rojas-Gutierrez, E.; Munoz-Arenas, G.; Trevino, S.; Espinosa, B.; Chavez, R.; Rojas, K.; Flores, G.; Diaz, A.; Guevara, J. Alzheimer's disease and metabolic syndrome: A link from oxidative stress and inflammation to neurodegeneration. *Synapse* **2017**, *71*, e21990. [CrossRef] [PubMed]
215. Guigoz, Y.; Dore, J.; Schiffrina, E.J. The inflammatory status of old age can be nurtured from the intestinal environment. *Curr. Opin. Clin. Nutr. Metab. Care* **2008**, *11*, 13–20. [CrossRef] [PubMed]
216. Garcia-Pena, C.; Alvarez-Cisneros, T.; Quiroz-Baez, R.; Friedland, R.P. Microbiota and aging: A review and commentary. *Arch. Med. Res.* **2017**, *48*, 681–689. [CrossRef]

217. Rojo, D.; Mendez-Garcia, C.; Raczkowska, B.A.; Bargiela, R.; Moya, A.; Ferrer, M.; Barbas, C. Exploring the human microbiome from multiple perspectives: Factors altering its composition and function. *FEMS Microbiol. Rev.* **2017**, *41*, 453–478. [CrossRef] [PubMed]
218. Turroni, F.; Ventura, M.; Butto, L.F.; Duranti, S.; O'Toole, P.W.; Motherway, M.O.C.; van Sinderen, D. Molecular dialogue between the human gut microbiota and the host: A Lactobacillus and Bifidobacterium perspective. *Cell. Mol. Life Sci.* **2014**, *71*, 183–203. [CrossRef]
219. Everard, A.; Belzer, C.; Geurts, L.; Ouwerkerk, J.P.; Druart, C.; Bindels, L.B.; Guiot, Y.; Derrien, M.; Muccioli, G.G.; Delzenne, N.M.; et al. Cross-talk between Akkermansia muciniphila and intestinal epithelium controls diet-induced obesity. *Proc. Natl. Acad. Sci. USA* **2013**, *110*, 9066–9071. [CrossRef]
220. Biagi, E.; Rampelli, S.; Turroni, S.; Quercia, S.; Candela, M.; Brigidi, P. The gut microbiota of centenarians: Signatures of longevity in the gut microbiota profile. *Mech. Ageing Dev.* **2017**, *165*, 180–184. [CrossRef]
221. Kogut, M.H.; Lee, A.; Santin, E. Microbiome and pathogen interaction with the immune system. *Poult. Sci.* **2020**, *99*, 1906–1913. [CrossRef]
222. Ruiz-Ruiz, S.; Sanchez-Carrillo, S.; Ciordia, S.; Mena, M.C.; Mendez-Garcia, C.; Rojo, D.; Bargiela, R.; Zubeldia-Varela, E.; Martinez-Martinez, M.; Barbas, C.; et al. Functional microbiome deficits associated with ageing: Chronological age threshold. *Aging Cell* **2020**, *19*, e13063. [CrossRef] [PubMed]
223. Ramos-Chavez, L.A.; Roldan-Roldan, G.; Garcia-Juarez, B.; Gonzalez-Esquivel, D.; Perez de la Cruz, G.; Pineda, B.; Ramirez-Ortega, D.; Munoz, I.G.; Herrera, B.J.; Rios, C.; et al. Low serum tryptophan levels as an indicator of global cognitive performance in nondemented women over 50 years of age. *Oxidative Med. Cell. Longev.* **2018**, *2018*, 8604718. [CrossRef] [PubMed]
224. Gao, J.; Xu, K.; Liu, H.; Liu, G.; Bai, M.; Peng, C.; Li, T.J.; Yin, Y.L. Impact of the gut microbiota on intestinal immunity mediated by tryptophan metabolism. *Front. Cell. Infect. Microbiol.* **2018**, *8*, e56564. [CrossRef]
225. Collino, S.; Montoliu, I.; Martin, F.P.J.; Scherer, M.; Mari, D.; Salvioli, S.; Bucci, L.; Ostan, R.; Monti, D.; Biagi, E.; et al. Metabolic signatures of extreme longevity in northern Italian centenarians reveal a complex remodeling of lipids, amino acids, and gut microbiota metabolism. *PLoS ONE* **2013**, *8*, e56564. [CrossRef]
226. Mace, J.L.; Porter, R.J.; Dalrymple-Alford, J.C.; Wesnes, K.A.; Anderson, T.J. Effects of acute tryptophan depletion on neuropsychological and motor function in Parkinson's disease. *J. Psychopharmacol.* **2010**, *24*, 1465–1472. [CrossRef] [PubMed]
227. Cisek, K.; Krochmal, M.; Klein, J.; Mischak, H. The application of multi-omics and systems biology to identify therapeutic targets in chronic kidney disease. *Nephrol. Dial. Transplant.* **2016**, *31*, 2003–2011. [CrossRef]
228. Montaner, J.; Ramiro, L.; Simats, A.; Tiedt, S.; Makris, K.; Jickling, G.C.; Debette, S.; Sanchez, J.C.; Bustamante, A. Multilevel omics for the discovery of biomarkers and therapeutic targets for stroke. *Nat. Rev. Neurol.* **2020**, *16*, 247–264. [CrossRef]
229. de Tayrac, M.; Le, S.; Aubry, M.; Mosser, J.; Husson, F. Simultaneous analysis of distinct Omics data sets with integration of biological knowledge: Multiple Factor Analysis approach. *BMC Genom.* **2009**, *10*, 32. [CrossRef]
230. West, L.; Vidwans, S.J.; Campbell, N.P.; Shrager, J.; Simon, G.R.; Bueno, R.; Dennis, P.A.; Otterson, G.A.; Salgia, R. A novel classification of lung cancer into molecular subtypes. *PLoS ONE* **2012**, *7*, e31906. [CrossRef]
231. Collisson, E.A.; Sadanandam, A.; Olson, P.; Gibb, W.J.; Truitt, M.; Gu, S.; Cooc, J.; Weinkle, J.; Kim, G.E.; Jakkula, L.; et al. Subtypes of pancreatic ductal adenocarcinoma and their differing responses to therapy. *Nat. Med.* **2011**, *17*, 500–503. [CrossRef]
232. Higdon, R.; Earl, R.K.; Stanberry, L.; Hudac, C.M.; Montague, E.; Stewart, E.; Janko, I.; Choiniere, J.; Broomall, W.; Kolker, N.; et al. The promise of multi-omics and clinical data integration to identify and target personalized healthcare approaches in autism spectrum disorders. *Omics—J. Integr. Biol.* **2015**, *19*, 197–208. [CrossRef] [PubMed]
233. Solovev, I.; Shaposhnikov, M.; Moskalev, A. Multi-omics approaches to human biological age estimation. *Mech. Ageing Dev.* **2020**, *185*, 111192. [CrossRef]
234. Ahadi, S.; Zhou, W.; Schussler-Fiorenza Rose, S.M.; Sailani, M.R.; Contrepois, K.; Avina, M.; Ashland, M.; Brunet, A.; Snyder, M. Personal aging markers and ageotypes revealed by deep longitudinal profiling. *Nat. Med.* **2020**, *26*, 83–90. [CrossRef]
235. Lopez-Otin, C.; Blasco, M.A.; Partridge, L.; Serrano, M.; Kroemer, G. The hallmarks of aging. *Cell* **2013**, *153*, 1194–1217. [CrossRef] [PubMed]
236. Mitteldorf, J. A clinical trial using methylation age to evaluate current antiaging practices. *Rejuvenation Res.* **2019**, *22*, 201–209. [CrossRef]
237. Soriano-Tarraga, C.; Giralt-Steinhauer, E.; Mola-Caminal, M.; Ois, A.; Rodriguez-Campello, A.; Cuadrado-Godia, E.; Fernandez-Cadenas, I.; Cullell, N.; Roquer, J.; Jimenez-Conde, J. Biological age is a predictor of mortality in ischemic stroke. *Sci. Rep.* **2018**, *8*, 4148. [CrossRef]
238. Cole, J.H.; Ritchie, S.J.; Bastin, M.E.; Hernandez, M.C.V.; Maniega, S.M.; Royle, N.; Corley, J.; Pattie, A.; Harris, S.E.; Zhang, Q.; et al. Brain age predicts mortality. *Mol. Psychiatry* **2018**, *23*, 1385–1392. [CrossRef]
239. Tat-Thang, V.; Vivot, A.; Porcher, R. Impact of biomarker-based design strategies on the risk of false-positive findings in targeted therapy evaluation. *Clin. Cancer Res.* **2018**, *24*, 6257–6264. [CrossRef]
240. Yao, M.; Wang, L.; Yao, Y.; Gu, H.B.; Yao, D.F. Biomarker-based microRNA therapeutic strategies for hepatocellular carcinoma. *J. Clin. Transl. Hepatol.* **2014**, *2*, 253–258. [CrossRef] [PubMed]
241. Blennow, K. Biomarkers in Alzheimer's disease drug development. *Nat. Med.* **2010**, *16*, 1218–1222. [CrossRef]
242. Balasubramanian, P.; Howell, P.R.; Anderson, R.M. Aging and caloric restriction research: A biological perspective with translational potential. *EBioMedicine* **2017**, *21*, 37–44. [CrossRef] [PubMed]