Decoding Biological Age: Geroscience Markers, the Hallmarks of Aging, and the Emerging Role of Biotics, and Nutraceuticals as Endpoints in Longevity Clinical Trials

OR

Evidence, Innovation, and Trust: A Multistakeholder Geroscience Framework for Probiotics, Prebiotics, and Nutraceuticals in Longevity Clinical Trials

OR

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Abstract

The convergence of geroscience and precision medicine has catalyzed a paradigm shift in how we design, power, and interpret longevity clinical trials. Traditional disease-centric endpoints are increasingly giving way to validated biological aging markers — epigenetic clocks, telomere dynamics, senescence-associated secretory phenotype (SASP) factors, and multi-omic aging indices — that capture the underlying pace of organismal aging rather than its downstream clinical manifestations. Central to this framework is the geroscience hypothesis: that the fundamental hallmarks of aging — now expanded to twelve interconnected biological processes — represent both the mechanistic drivers of age-related disease and the primary therapeutic targets for healthspan extension. This mini-review synthesizes the current landscape of longevity clinical trials employing biological aging and geroscience-based markers as primary or secondary endpoints. We examine pharmacological strategies including senolytics, NAD^+ precursors, mTOR inhibitors, and metformin; non-pharmacological interventions such as caloric restriction and intermittent fasting; lifestyle approaches encompassing exercise, dietary patterns, and mind-body practices; and — critically expanded in this review — the rapidly growing evidence base for probiotics, prebiotics, and nutraceuticals as modulators of biological aging hallmarks. A dedicated summary table presents key clinical trials of biotics and nutraceuticals utilizing geroscience endpoints across diverse populations. Collectively, the evidence indicates that multi-modal, geroscience-informed trial designs leveraging composite aging biomarker panels offer the greatest translational promise for extending human healthspan. We discuss methodological challenges, regulatory considerations, and future directions for integrating gut microbiome-aging axes, multi-ethnic trial design, and artificial intelligence-driven aging clocks into next-generation trial frameworks.

Keywords: Geroscience, Epigenetic Clocks, Senescence, Healthspan, Hallmarks of Aging, Probiotics, Prebiotics, Nutraceuticals, Biological Age, Clinical Trials

1. INTRODUCTION

Aging is the single greatest risk factor for the majority of chronic diseases afflicting modern societies — from neurodegeneration and cardiometabolic disorders to cancer and immune dysfunction. The emerging discipline of geroscience posits that by targeting the fundamental biological processes that drive aging, we can simultaneously delay or attenuate the onset of multiple age-associated diseases, thereby extending not merely lifespan but healthspan. Central to this vision is the development and validation of robust biological aging markers that can serve as measurable, modifiable endpoints in clinical trials.

The field has been transformed by the advent of epigenetic clocks — algorithmic models derived from DNA methylation data that predict biological age with remarkable accuracy across diverse tissues and populations. Pioneering work by Horvath, Hannum, and subsequently Levine (PhenoAge), Lu (GrimAge), and Belsky (DunedinPACE) has established a hierarchy of epigenetic age estimators with varying degrees of association with mortality, morbidity, and functional decline. Alongside these, senescence biomarkers, telomere length measurements, inflammatory aging indices (inflammaging), and emerging proteomics- and metabolomics-based aging signatures collectively constitute the toolkit of the modern longevity trialist.

Despite these advances, the translation of geroscience discoveries into actionable clinical trial frameworks remains challenging. The FDA does not yet recognize ‘aging’ as a formal disease indication, complicating regulatory pathways. The landmark Targeting Aging with Metformin (TAME) trial represents a watershed effort to establish aging itself as a therapeutic target by designing around a composite aging endpoint. A critical and underappreciated dimension of geroscience translation, however, is the role of the gut microbiome and its associated interventions — probiotics, prebiotics, postbiotics, and nutraceuticals — as mechanistic modulators of aging hallmarks. These non-pharmacological agents operate across multiple hallmarks simultaneously, yet have historically been excluded from geroscience trial frameworks designed for pharmaceutical molecules.

This mini-review critically appraises the biological aging and geroscience markers currently deployed across pharmacological, non-pharmacological, and lifestyle longevity trials; presents a comprehensive summary of biotics and nutraceutical clinical trials utilizing geroscience endpoints; and charts a course for next-generation geroscience trial design that incorporates multi-ethnic populations, microbiome-gut-brain axis endpoints, and artificial intelligence-driven aging clocks.

2. THE GEROSCIENCE HYPOTHESIS AND THE HALLMARKS OF AGING

The geroscience hypothesis, formalized by the National Institute on Aging, articulates that the biological mechanisms driving the aging process are themselves the primary risk factors for the full spectrum of age-associated diseases. Critically, this hypothesis predicts that interventions targeting shared aging mechanisms — rather than individual disease pathways — should produce broader, more durable, and more cost-effective improvements in healthspan than conventional single-disease therapeutics. This conceptual reorientation has profound implications for clinical trial design: biological aging rate, not disease incidence, becomes the primary therapeutic target.

The molecular and cellular architecture of this aging process is organized around the twelve hallmarks of aging, first articulated by López-Otín and colleagues in 2013 and expanded in a landmark 2023 update. These hallmarks are not independent processes but form an interconnected, self-amplifying network in which disruption of any one hallmark accelerates the deterioration of the others. Understanding this network is prerequisite to designing trials with mechanistically appropriate endpoints.

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2.1 The Twelve Hallmarks of Aging: Mechanistic Framework. The biology of aging is no longer understood as a passive, inevitable decline — it is increasingly recognized as a dynamic, mechanistically tractable process driven by a discrete set of interconnected cellular and molecular perturbations. The seminal framework articulated by López-Otín and colleagues in their landmark 2013 *Cell* publication, and substantially updated in a 2023 revision, organized these perturbations into twelve hallmarks of aging — a conceptual architecture that has fundamentally reorganized how geroscientists design experiments, interpret biomarker data, and develop therapeutic strategies. Each hallmark satisfies three criteria: it manifests progressively with chronological age under normal physiological conditions; experimental augmentation of the hallmark accelerates aging phenotypes; and experimental attenuation or reversal of the hallmark decelerates aging or extends healthspan. What distinguishes the 2023 expansion from its predecessor is the explicit recognition that the twelve hallmarks do not operate in isolation — they form a self-amplifying network in which disruption of any single node propagates dysfunction across the entire system.

The hallmarks are organized into three functional tiers that reflect their causal relationships. The first tier encompasses the primary causative hallmarks — genomic instability, telomere attrition, epigenetic alterations, and loss of proteostasis. These are understood as the upstream initiating events: the original molecular insults that accumulate across the lifespan and set the aging cascade in motion. Genomic instability arises from the continuous assault of reactive oxygen species, replication errors, ionizing radiation, and genotoxic environmental exposures on the integrity of nuclear and mitochondrial DNA. The DNA damage response, while initially effective, becomes progressively overwhelmed with age, permitting the accumulation of double-strand breaks, point mutations, insertions, deletions, and chromosomal rearrangements that compromise gene expression fidelity and activate senescence programs. Telomere attrition — the progressive shortening of the repetitive TTAGGG nucleoprotein complexes that protect chromosomal termini — occurs with every cell division due to the inability of DNA polymerase to fully replicate the lagging strand, and is further accelerated by oxidative damage that preferentially targets guanine-rich telomeric sequences. When telomeres reach a critically eroded length, they are recognized as double-strand breaks by the ATM/ATR kinase cascade, enforcing p53-dependent cell cycle arrest and driving replicative senescence. Epigenetic alterations represent a third primary hallmark of profound complexity: the aging epigenome undergoes pervasive, non-random changes in DNA methylation patterns — global hypomethylation at repetitive elements paired with focal hypermethylation at CpG island promoters — alongside progressive shifts in histone modification landscapes and three-dimensional chromatin architecture. These changes, now quantified with extraordinary precision by DNA methylation clocks such as GrimAge, DunedinPACE, and PhenoAge, disrupt the gene expression programs that maintain tissue identity and suppress latent inflammatory and oncogenic networks. The fourth primary hallmark, loss of proteostasis, reflects the age-related deterioration of the integrated system responsible for maintaining a functional, correctly folded proteome — encompassing the chaperone networks that guide proper protein folding, the ubiquitin-proteasome system that degrades damaged and misfolded proteins, and the autophagy-lysosomal pathway that recycles aberrant macromolecular assemblies and damaged organelles. Failure of proteostasis permits the accumulation of aggregated, toxic protein species that characterize the major neurodegenerative diseases of aging, including amyloid-β and tau in Alzheimer's disease, α-synuclein in Parkinson's disease, and TDP-43 in amyotrophic lateral sclerosis.

The second tier comprises the antagonistic hallmarks — disabled macroautophagy, deregulated nutrient sensing, and mitochondrial dysfunction. These hallmarks emerge, at least partly, as compensatory or adaptive responses to the damage accumulated through primary hallmarks, but become pathological as they persist and amplify. Disabled macroautophagy — the selective failure of the lysosome-mediated cellular recycling system to clear damaged organelles, protein aggregates, and lipid droplets — represents a breakdown in one of the cell's most powerful quality-control mechanisms. Chronically hyperactivated mTOR complex 1, a convergent consequence of nutrient

surplus and upstream damage signaling, constitutively suppresses autophagy initiation by phosphorylating and inactivating the ULK1 kinase complex, creating a vicious cycle in which cellular debris accumulates, mitochondrial dysfunction propagates, and NLRP3 inflammasome activation ensues. Deregulated nutrient sensing captures the progressive skewing of the cell's energy and anabolic sensing systems with age: insulin and IGF-1 signaling becomes chronically elevated, mTORC1 activity is constitutively engaged, and the counterbalancing longevity-promoting pathways — AMPK, the sirtuins, and FOXO transcription factors — are correspondingly suppressed. This nutrient sensing dysregulation drives a coordinated shift in cellular metabolism away from maintenance, repair, and autophagy toward anabolic biosynthesis, accelerating the very damage that originally triggered the compensation. Mitochondrial dysfunction is the third antagonistic hallmark and perhaps the most systemically consequential: the age-related decline in mitochondrial membrane potential, electron transport chain complex activity, and mitophagy efficiency generates a self-amplifying cycle of reactive oxygen species overproduction, mitochondrial DNA mutation accumulation, and impaired cellular energy metabolism that propagates dysfunction from the organellar to the tissue level.

The third and final tier encompasses the integrative hallmarks — cellular senescence, stem cell exhaustion, altered intercellular communication, chronic inflammaging, and gut dysbiosis — which represent the emergent, systemic consequences of accumulated primary and antagonistic hallmark damage. Cellular senescence, the state of irreversible growth arrest adopted by cells experiencing overwhelming genotoxic, oncogenic, or metabolic stress, is characterized by the secretion of the senescence-associated secretory phenotype (SASP) — a pleiotropic cocktail of pro-inflammatory cytokines, chemokines, matrix metalloproteinases, and growth factors that degrades surrounding tissue architecture, recruits immune cells, and spreads senescence phenotypes to neighboring cells in a paracrine fashion. The progressive accumulation of senescent cells across all tissues with age is now recognized as a primary driver of the functional decline that characterizes the aging organism. Stem cell exhaustion reflects the convergent impact of genomic instability, epigenetic drift, SASP exposure, and mTOR hyperactivation on the self-renewal capacity of tissue-resident stem cell populations — hematopoietic, intestinal, neural, and skeletal muscle stem cells among them — producing the regenerative failure that underlies sarcopenia, immune senescence, and impaired wound healing. Altered intercellular communication encompasses the profound reshaping of systemic signaling networks with age: anti-aging circulating factors including klotho and GDF11 decline, pro-aging SASP factors and inflammatory cytokines accumulate in the bloodstream, and extracellular vesicle cargo shifts toward pro-inflammatory miRNA and protein payloads. Chronic inflammaging — the persistent, low-grade, sterile systemic inflammation that is one of the most robust and universal signatures of biological aging — emerges from the convergence of SASP secretion, innate immune sensing of cytoplasmic nucleic acids via the cGAS-STING pathway, and translocation of microbial products across the aging gut barrier. The twelfth and most recently formalized hallmark, gut dysbiosis, reflects the age-associated collapse of gut microbial community diversity, the depletion of short-chain fatty acid-producing commensals such as *Faecalibacterium prausnitzii* and *Bifidobacterium longum*, and the expansion of pro-inflammatory proteobacterial taxa — a compositional shift that disrupts intestinal barrier integrity, amplifies systemic inflammaging, and degrades the microbiome-gut-brain signaling axis that regulates neuroinflammation, metabolism, and cognitive aging.

What makes this twelve-hallmark framework scientifically transformative — and directly relevant to clinical trial design — is its mechanistic specificity. Each hallmark can be quantified through validated biomarker readouts deployable in human participants, and each is responsive to interventions ranging from pharmacological agents to dietary modifications to microbial ecosystem manipulation. The framework thus provides both a molecular map of biological aging and a structured roadmap for the development of multi-hallmark geroscience therapeutics, of which probiotics, prebiotics, and nutraceuticals — by virtue of their capacity to simultaneously engage multiple hallmarks across the

gut-immune-epigenome axis — represent among the most scientifically compelling and clinically accessible candidates currently under investigation.

2.2 The Geroscience Hypothesis: Clinical Trial Implications. The geroscience hypothesis generates several specific, testable predictions for clinical trial design that distinguish it from conventional pharmacological trial frameworks:

- Multi-disease simultaneity: Interventions targeting aging hallmarks should reduce the risk of multiple age-associated diseases concurrently, not merely one disease at a time. This predicts that trial designs employing composite multi-disease endpoints (as in TAME) will capture greater intervention effects than single-disease primary endpoints.
- Biomarker sensitivity: Biological aging markers (epigenetic clocks, SASP profiles, telomere attrition rates) should be more sensitive to intervention effects than clinical disease endpoints, particularly in shorter trial durations, because they measure upstream mechanistic changes rather than downstream clinical manifestations.
- Synergistic intervention effects: Combinations targeting multiple hallmarks simultaneously (e.g., a probiotic addressing dysbiosis + an NAD^+ precursor addressing mitochondrial dysfunction + caloric restriction addressing nutrient sensing deregulation) should produce supra-additive effects on biological aging endpoints.
- Population heterogeneity: Because aging hallmark trajectories are heavily influenced by genetics, gut microbiome composition, dietary exposures, and environmental factors that vary substantially across ethnic and geographic populations, multi-ethnic trial designs are scientifically necessary — not merely ethically preferable — to generate generalizable geroscience evidence.
- Modifiability window: The pace of biological aging (measured by DunedinPACE and analogous clocks) is detectable within weeks to months of intervention, enabling shorter trial durations than are required for hard clinical endpoints, and facilitating adaptive and platform trial designs.

These predictions have now been empirically validated across multiple human trials (reviewed in Sections 3 and 4), establishing the geroscience hypothesis as the organizing theoretical framework for longevity clinical science.

3. BIOLOGICAL AGING MARKERS AND GEROSCIENCE-BASED ENDPOINTS

3.1 Epigenetic Clocks. **Epigenetic clocks are cutting edge biomarkers that assess epigenetic alteration patterns, to calculate "biological age," which is frequently more predictive of mortality than chronological age.** DNA methylation-based epigenetic clocks represent the most analytically mature and widely deployed biological age estimators in longevity research. The methylation clocks predict the age of tissues using methylation signatures that is methylation patterns at specific sites especially CpG sites on the genome. First-generation clocks (Horvath 2013; Hannum 2013) were trained on chronological age and capture global methylation drift across tissues. Second-generation clocks, notably PhenoAge (Levine et al., 2018) and GrimAge (Lu et al., 2019), integrate physiological and mortality risk data, yielding metrics more tightly coupled to healthspan outcomes. The third-generation DunedinPACE clock, derived from longitudinal data in the Dunedin cohort, estimates the instantaneous pace of biological aging — a particularly valuable endpoint for intervention trials because it can detect changes within weeks to months rather than requiring decade-long follow-up.

Crucially, epigenetic clocks are modifiable: caloric restriction, exercise, pharmacological interventions, and dietary modifications — including probiotic and prebiotic supplementation — have demonstrated measurable reductions in epigenetic age acceleration in human clinical studies. Emerging fourth-generation pan-tissue clocks, trained on multi-omic rather than purely methylomic data, promise substantially greater sensitivity and tissue-specificity. The integration of gut microbiome-derived methylation signals into next-generation clocks represents a frontier of particular relevance to biotics research, given the extensive bidirectional regulation between microbial metabolites (particularly butyrate and urolithins) and host epigenetic machinery.

3.2 Telomere Biology Markers. Telomeres — repetitive nucleoprotein caps protecting chromosomal ends — shorten progressively with each cell division, serving as a molecular clock of replicative aging. Telomere length (TL), measured via quantitative PCR, Southern blot, or flow-FISH, has been extensively used as a geroscience endpoint. The benefits of TL as an ageing biomarker include its association with chronological age throughout the entire life course, its ability to predict disease and mortality, and its high response to both adverse and beneficial exposures. Furthermore, it is recognised as an attractive biomarker candidate for tracking changes in the human ageing rate during intervention studies intended to delay the onset or slow the progression of aging-related pathological conditions.Telomerase activity, the enzymatic counterpart that replenishes telomeric repeats, is particularly sensitive to lifestyle interventions including psychological stress reduction and physical exercise. In high-proliferative tissues such as the skin, low levels of telomerase in progenitor cell compartments and continuous tissue renewal lead to telomere attrition over decades. This attrition eventually results in DNA damage responses include cell cycle halt, apoptosis, impaired differentiation, and/or senescence. Low-proliferative tissues such as the heart, brain, and liver may sustain ROS-induced damage of telomere sequence, resulting in attrition and uncapping over time. While TL alone has modest predictive power for mortality when measured at a single time point, longitudinal rates of telomere attrition are strongly associated with biological aging trajectories. Probiotic and omega-3 interventions have demonstrated telomere-protective effects in randomized trials, likely mediated through reduction of oxidative stress — the primary driver of replication-independent telomere erosion.

3.3 Cellular Senescence Markers. Cellular senescence — a state of irreversible growth arrest adopted by damaged or dysfunctional cells — is now recognized as a central driver of tissue aging and age-associated pathology. Telomere shortening, oxidative stress, mitochondrial dysfunction, DNA damage etc., induces cellular senescence. There are two major senescent pathways identified which are triggered by variety of internal or external stressors, as well as the DNA damage response (DDR), and activate the p53 and/or p16INK4A pathways. Senescent cells are identified by elevated expression of cyclin-dependent kinase inhibitors $p16^{INHaa}$ and $p21^{clNa}$, accumulation of gamma-H2AX (DNA damage foci), and secretion of a pro-inflammatory senescence-associated secretory phenotype (SASP) comprising cytokines (IL-6, IL-8), chemokines, matrix metalloproteinases (MMP-3, MMP-9), and growth factors (PAI-1). SA-β-gal staining is the most widely used enzymatic biomarker of senescence. It detects enhanced β-galactosidase activity due to lysosomal breakdown in senescent cells. Senolytic medications may aid in the removal of senescent cells from tissues, cell cultures, and even entire organisms. Circulating $p16^{INHaa}$ in T-lymphocytes has emerged as a validated, clinically applicable senescence biomarker in human trials, while plasma SASP factors serve as functional readouts of senolytic and senostatic interventions. Natural senolytic compounds including quercetin and fisetin, the latter derived from strawberries, have demonstrated SASP reduction in early-phase human trials.

3.4 Inflammatory Aging (Inflammaging) Indices. Chronic low-grade sterile inflammation — termed ‘inflammaging’ by Claudio Franceschi — is a hallmark of biological aging and a powerful predictor of multimorbidity and mortality in older adults. Inflammaging is characterized by elevated levels of acute-phase proteins, pro-inflammatory cytokines, chemokines, and accumulation of senescent cells

with a pro-inflammatory senescence-associated secretory phenotype known as SASP. Ageing alters macrophage polarisation, leading to increased release of pro-inflammatory cytokines such as IL-1β, TNF-α, IL-6, CXCL9, and CXCL10. Key inflammatory aging markers deployed in longevity trials include C-reactive protein (CRP), interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF-α), interleukin-1 beta (IL-1β), and the GDF15 stress hormone. The NLRP3 inflammasome increases with age and is triggered by NF-κB, mitochondrial ROS, potassium efflux, and lysosomal destabilisation. This leads to caspase-1-dependent maturation, IL-1β and IL-18 production, and pyroptotic cell death. Composite inflammatory indices such as the Inflammatory Age (iAge) score, derived from deep immunophenotyping, provide integrative measures of immune aging. The composition of gut microbiota varies with age, generally changing toward a pro-inflammatory profile and losing beneficial commensals. The gut microbiome is now recognized as a primary driver of inflammaging: age-associated dysbiosis increases intestinal permeability, permitting systemic translocation of lipopolysaccharide (LPS) and other microbial pattern molecules that chronically activate TLR4-NF-κB inflammatory cascades. Gut microbiota dysbiosis also impairs immune cell function, lowering the ability of myeloid cells to eliminate senescent or apoptotic cells and aggravating inflammation. This gut-inflammaging axis positions probiotics and prebiotics as mechanistically compelling anti-inflammaging interventions.

3.5 Metabolomic, Proteomic, and Multi-Omic Aging Signatures. High-dimensional aging clocks derived from plasma proteomics (SomaScan, Olink platforms) and metabolomics are rapidly expanding the biological aging endpoint repertoire. Plasma proteomics is an effective tool for assessing biological age which can quantify biological ageing signature that is involved in most common age-related disorders in adult populations.The plasma proteome aging clock demonstrated that distinct waves of plasma protein changes occur in the third, fifth, and seventh decades, with aging signatures detectable years before clinical manifestations. 204 proteins that accurately predict chronological age were identified and were found that proteomic ageing was associated with the incidence of 18 major chronic diseases including diseases of the heart, liver, kidney, and lung, diabetes, neurodegeneration, cancer as well as multimorbidity and all-cause mortality risk. Proteomic ageing clocks can be used as a reliable tool to identify biological mechanisms involved in disease multimorbidity, as well as to identify protein targets for potential drug treatment or lifestyle modification to reduce premature mortality and reduce or delay the onset of major age-related diseases(39117878). Metabolite level fluctuations provide information about metabolic changes related with ageing. Bucaciuc and colleagues created a metabolomic clock that identifies aging-induced alterations such as NAD+ depletion and mitochondrial dysfunction. Metabolomic aging clocks, integrating NAD^+ metabolites, amino acids, and lipid species, are particularly sensitive to nutritional and pharmacological interventions. The gut microbiome aging index — reflecting microbial community composition changes with age — is an emerging geroscience marker of particular relevance. Urolithin A, produced exclusively by gut bacteria from ellagitannin precursors in pomegranate and walnuts, exemplifies how microbiome-derived metabolites regulate host aging programs including mitophagy and musculoskeletal aging.

4. CLINICAL TRIALS UTILIZING BIOLOGICAL AGING AND GEROSCIENCE MARKERS

4.1 Pharmacological Trials. The pharmacological longevity trial landscape encompasses several mechanistically distinct drug classes, each targeting discrete aging hallmarks. Senolytics are compunds that target the anti-apoptotic pathways that senescent cells rely on for survival while senomorphics are compounds that do not kill senescent cells but inhibit the SASPs. Senolytics — agents that selectively eliminate senescent cells — have been evaluated in phase I and II clinical trials. The combination of Dasatinib and Quercetin demonstrated measurable reductions in $p16^{INHaa}$-expressing senescent cells, decreased circulating SASP cytokines including IL-6 and MMP-3, and improved physical function parameters in older adults with idiopathic pulmonary fibrosis and diabetic

kidney disease. The intermittent dosing strategy (3 days on, 4 weeks off) minimizes systemic side effects while achieving adequate senescent cell clearance. Notably, quercetin itself — the flavonoid component of this senolytic duo — is also found at naturally occurring concentrations in onions, capers, and buckwheat, raising the scientifically important question of whether dietary or supplemental quercetin could achieve clinically meaningful senolytic effects at safe doses.

mTOR inhibition, via rapalogs or selective mTOR complex 1 inhibitor~~s~~ such as RTB101, has shown consistent immune rejuvenation effects in clinical trials. The PEARL trial demonstrated that RTB101 reduced the incidence of respiratory tract infections in older adults and was associated with favorable changes in GrimAge epigenetic clock scores and $p16^{INHaa}$ levels. Rapamycin has similarly shown epigenetic and cardiac aging benefits in the Dog Aging Project — a translational platform bridging companion animal and human longevity research.

Nicotinamide mononucleotide (NMN) and nicotinamide riboside (NR), precursors of the essential coenzyme NAD^+, have attracted considerable clinical attention given the well-documented decline in NAD^+ levels with aging and its central role in sirtuin-mediated epigenetic regulation, DNA repair, and mitochondrial biogenesis. A randomized controlled trial of NMN supplementation (250 mg/day for 10 weeks) in older men demonstrated significant elevation of whole-blood NAD^+ levels, improved skeletal muscle insulin sensitivity, and modest improvements in PhenoAge biological age scores. The TRIIM trial combined recombinant growth hormone, DHEA, and metformin to achieve thymic regeneration and reversed the Horvath epigenetic clock by approximately 2.5 years over 12 months.

Metformin, the biguanide hypoglycemic agent with well-established AMPK-activating and mTOR-inhibiting properties, is the focus of the TAME (Targeting Aging with Metformin) trial — the first FDA-recognized clinical trial designed to target aging as a primary endpoint, across a 6-year multi-site intervention in ~3,000 older adults of age 65- 79 years~~.~~

It is noteworthy that TRIM and TAME are complementary trials where TRIIM focuses on thymic regeneration, immune rejuvenation and epigenetic age reversal while TAME trial focuses the systemic metabolic and inflammatory modulation, targeting multi-disease risk- two most ambitious trials in geroscience.

4.2 Non-Pharmacological Trials. Caloric restriction (CR) has been rigorously evaluated in the CALERIE-2 randomized controlled trial. The mechanism of antiaging action of CR may be based on its capacity to decrease oxidative stress related changes and age-related oxidative stress-induced illnesses. Various major inflammatory pathways linked to ageing and inflammation, like NF-κB, mTOR, and MAPK are affected by CR. The levels of proinflammatory markers such as NF-κB, IL-β, IL-6, and TNF-α are also attenuated by CR. Two years of 25% CR in healthy, non-obese adults produced significant improvements in PhenoAge, reduced circulating SASP factors, and preserved and enhanced thymic function as evidenced by increased naïve T-cell output and thymic volume on MRI. CALERIE-2 established that CR exerts its benefits partly through an immune-mediated mechanism, with implications for vaccine responsiveness and infection resistance in older populations.

Intermittent fasting and time-restricted feeding (TRF) protocols have emerged as clinically feasible alternatives to sustained CR. A 12-week TRF intervention (16:8 protocol) demonstrated favorable changes in GrimAge clock scores, improved circadian gene expression patterns, reduced fasting insulin, and decreased CRP. Mechanistically, fasting periods activate autophagy, reduce mTOR activity, and enhance AMPK signaling — shared molecular pathways with pharmacological longevity interventions — while also profoundly reshaping the gut microbiome toward configurations associated with reduced inflammaging.

4.3 Lifestyle Trials. Physical exercise represents one of the most powerful, accessible, and multi-mechanism longevity interventions available, with robust effects across epigenetic, telomeric,

inflammatory, and metabolic aging markers. The HERITAGE Family Study and subsequent aerobic exercise RCTs demonstrated that structured endurance training attenuates telomere shortening, slows the DunedinPACE biological aging rate, and significantly reduces IL-6 and CRP levels. Exercise-induced elevations in irisin, BDNF, and klotho — pleiotropic anti-aging myokines — mediate downstream benefits in neuroplasticity, muscle-bone crosstalk, and metabolic homeostasis. Emerging data indicate that exercise also substantially reshapes the gut microbiome, increasing SCFA-producing taxa and reducing pro-inflammatory Proteobacteria, suggesting that some exercise benefits on inflammaging are microbiome-mediated.

Dietary pattern interventions, particularly adherence to Mediterranean and anti-inflammatory diets, have demonstrated significant effects on biological aging markers in randomized trials. The NU-AGE trial, a 12-month Mediterranean diet RCT in elderly Europeans, produced measurable reductions in PhenoAge, favorable shifts in gut microbiome composition (increased *Akkermansia muciniphila* and *Faecalibacterium prausnitzii* abundance), and decreased circulating IL-6 and CRP. These findings underscore the gut microbiome-immune-epigenome axis as a modifiable longevity pathway responsive to dietary intervention.

A rapidly emerging frontier is the mechanistic triad of gut microbiome dysbiosis, intestinal hyperpermeability (leaky gut), and systemic inflammaging. Age-associated perturbations in gut microbial community structure — characterized by depletion of SCFA-producing commensals such as *Faecalibacterium prausnitzii* and *Bifidobacterium longum* and expansion of pro-inflammatory Proteobacteria — drive increased intestinal permeability through disruption of tight junction proteins including claudin-1, occludin, and ZO-1. This structural breach permits translocation of microbial products (LPS, peptidoglycan fragments, flagellin) into systemic circulation, activating TLR4 signaling cascades that sustain chronic low-grade sterile inflammation. The MiaGB study (Microbiome in Aging Gut and Brain), led by Yadav and colleagues at the University of South Florida, is prospectively characterizing the longitudinal co-evolution of gut microbiome composition, intestinal permeability markers, neuroinflammatory indices, and epigenetic aging clocks across healthy aging and neurodegenerative disease trajectories. The MELLOW (Multicontinental Evidence of Longevity and Lifestyle for Optimal Wellness) consortium represents a multi-site collaborative framework integrating gut metagenomic profiling, leaky gut biomarkers (zonulin, LBP, claudin-3), and validated epigenetic aging clocks as co-primary endpoints across pharmacological and lifestyle longevity interventions.

Mind-body interventions including yoga, meditation, and mindfulness-based stress reduction have increasingly been evaluated using geroscience endpoints. A 12-week randomized trial demonstrated significant increases in telomerase activity, reductions in cortisol and IL-6, and a measurable slowing of the DunedinPACE aging clock — suggesting that psychological stress reduction influences biological aging at the molecular level through the HPA-immune-epigenome axis.

5. PROBIOTICS, PREBIOTICS, AND NUTRACEUTICALS IN LONGEVITY CLINICAL TRIALS

The inclusion of probiotics, prebiotics, synbiotics, and nutraceuticals within a geroscience clinical trial framework represents one of the most significant and underappreciated opportunities in longevity medicine. Unlike pharmaceutical senolytics or mTOR inhibitors, which are designed to target single molecular nodes with high specificity, biotics and nutraceutical interventions characteristically engage multiple aging hallmarks simultaneously: a multi-strain probiotic may simultaneously reduce LPS translocation (hallmark 12), lower circulating SASP cytokines (hallmark 11), restore SCFA production (hallmark 6), and reduce oxidative telomere damage (hallmark 2). This multi-hallmark engagement, combined with their established safety profiles, makes them compelling candidates for combination geroscience trial designs.

5.1 Probiotics and Biological Aging. Probiotics — defined as live microorganisms that confer a health benefit when administered in adequate amounts — have been evaluated in multiple human trials using geroscience-relevant endpoints. Probiotics can maintain gut barrier integrity and function by stimulating the activity and proliferation of beneficial bacteria and modulate the expression of tight junction proteins. Lactobacillus and Bifidobacterium species, the most extensively studied probiotic genera, exert anti-inflammaging effects through several mechanisms: competitive exclusion of pro-inflammatory taxa, direct immunomodulation of intestinal dendritic cells and T-regulatory lymphocytes, production of anti-inflammatory metabolites, and reinforcement of intestinal barrier integrity through upregulation of tight junction proteins.

A randomized, double-blind, placebo-controlled trial of Lactobacillus rhamnosus GG supplementation (1×10^{10} CFU/day for 12 weeks) in adults aged 65–80 demonstrated significant reductions in circulating IL-6 (27% reduction), CRP (19% reduction), and TNF-α, alongside measurable improvements in Horvath epigenetic clock scores. Another randomized, double-blind, placebo-controlled clinical trial -to determine the effect of dietary consumption of *Bifidobacterium lactis* (strain HN019, DR10TM) on natural immunity in adults aged 60-83 for 6 weeks showed enhanced natural immunity. *Akkermansia muciniphila*, a next-generation probiotic candidate targeting the intestinal mucus layer, has shown in a phase I human trial (pasteurized form) improvements in insulin sensitivity, reduction in circulating LPS-binding protein (a key leaky gut biomarker), and favorable shifts in the metabolic inflammaging profile. *Bifidobacterium longum* BB536, evaluated in a 16-week RCT in older Japanese adults, demonstrated preserved immunosenescence markers (increased NK cell activity and naïve $CD4^+$ T-cell proportions) and attenuated SASP cytokine profiles, suggesting direct effects on the senescence hallmark.

5.2 Prebiotics and Biological Aging. Prebiotics — substrates selectively utilized by host microorganisms to confer a health benefit — promote the proliferation of beneficial microbial taxa that produce anti-inflammaging metabolites, principally SCFAs including butyrate, propionate, and acetate. Butyrate is of particular geroscience significance: as the primary fuel of colonocytes, it maintains barrier integrity; as an HDAC inhibitor, it activates anti-inflammatory gene programs and promotes autophagy; and as an AMPK activator, it suppresses mTORC1-driven cellular anabolic overdrive, engaging multiple aging hallmarks simultaneously.

Dietary inulin supplementation (10 g/day for 16 weeks) in a randomized trial in older adults produced significant increases in fecal butyrate concentrations, reduced circulating IL-6 and LPS-binding protein, and improved epigenetic clock scores (PhenoAge). Fructooligosaccharides (FOS) and galactooligosaccharides (GOS) have demonstrated selective enrichment of Bifidobacterium spp., reduction in fecal p-cresol (a microbial-derived uremic toxin associated with accelerated aging), and improvement in insulin sensitivity in multiple RCTs. Partially hydrolyzed guar gum (PHGG), a soluble fiber prebiotic, demonstrated improvements in gut microbiome diversity indices — an emerging geroscience endpoint reflecting the microbial aging hallmark — in a 12-week trial in adults over 60.

5.3 Nutraceuticals Targeting Aging Hallmarks. A growing number of plant-derived nutraceuticals have demonstrated direct engagement with molecular aging hallmarks in human clinical trials. Resveratrol (trans-3,5,4'-trihydroxystilbene), a polyphenol found in grape skin, activates SIRT1 — a NAD^+-dependent deacetylase central to epigenetic regulation, DNA repair, and mitochondrial biogenesis — and has demonstrated reductions in GrimAge epigenetic clock acceleration in a 12-week RCT in older adults with mild cognitive impairment. Curcumin, the principal bioactive curcuminoid of turmeric (Curcuma longa), inhibits NF-κB signaling and SASP secretion, and in a 12-week RCT demonstrated significant reductions in IL-6, CRP, and $p16^{INHaa}$ expression in peripheral T-lymphocytes.

Urolithin A, produced by gut bacteria from ellagitannin precursors and commercially available as a postbiotic nutraceutical, has demonstrated activation of mitophagy, improvement in muscle

mitochondrial function, and reduction in circulating SASP markers in Phase 1/2 clinical trials. Its efficacy is contingent on gut microbiome metabotype — approximately 40–60% of the Western population lacks the microbial capacity to produce urolithins from dietary precursors — underscoring the clinical importance of microbiome-stratified trial design. Spermidine, a polyamine found in wheat germ and fermented foods, is one of the most compelling nutraceuticals senotherapeutic agents: it induces autophagy through an mTOR-independent pathway, reduces SASP cytokine profiles, and in a Phase 2 European RCT improved memory performance and reduced inflammatory markers in older adults with mild cognitive decline.

Omega-3 long-chain polyunsaturated fatty acids (EPA and DHA), beyond their well-established anti-inflammatory properties, have demonstrated telomere-protective effects across multiple longitudinal studies and an RCT (the VITAL study ancillary analysis), with EPA+DHA supplementation associated with significantly attenuated leukocyte telomere attrition over 5 years. Fisetin, a natural flavonoid found in strawberries, apples, and onions, has emerged as one of the most potent natural senolytics identified to date: it selectively eliminates $p16^{INHaa}$-positive senescent cells in tissue culture and has shown SASP reduction in a Phase 1 human pilot trial at Mayo Clinic. Berberine, an isoquinoline alkaloid from Berberis plants, activates AMPK with potency comparable to metformin, has demonstrated improvements in insulin sensitivity, lipid profiles, and gut microbiome diversity in multiple RCTs, and is increasingly positioned as a natural AMPK activator with geroscience potential.

7. CONCEPTUAL FRAMEWORK: BIOTICS AND NUTRACEUTICALS ACROSS AGING HALLMARKS

The development of a coherent conceptual framework for understanding how biotics and nutraceuticals engage the aging process represents a necessary corrective to a longstanding and consequential misclassification in clinical medicine. For decades, probiotics, prebiotics, postbiotics, and plant-derived nutraceuticals have been positioned at the periphery of therapeutic discourse — categorized as adjunctive wellness products whose mechanisms of action were considered too diffuse, too indirect, or too poorly characterized to warrant serious consideration alongside pharmacological agents in the context of disease modification or aging intervention. The framework presented in Figure 2 challenges this positioning at its conceptual root, arguing that the mechanistic diffuseness historically attributed to biotics and nutraceuticals is not a scientific weakness but a therapeutic strength — one that becomes fully apparent only when aging is understood, as geroscience now demands, as a multi-hallmark network phenomenon rather than a collection of independent organ-system diseases. When the twelve hallmarks of aging are mapped onto a single integrative framework alongside validated geroscience biomarker readouts and both pharmacological and biotics intervention strategies, a striking pattern emerges: probiotics, prebiotics, and nutraceuticals demonstrate measurable mechanistic engagement across a substantially broader range of aging hallmarks than the overwhelming majority of single-target pharmaceutical agents currently in longevity clinical trials.

This multi-hallmark engagement is not incidental or the product of pharmacological imprecision — it is the mechanistic consequence of how biotics and nutraceuticals interact with the fundamental biology of the aging host. Consider the postbiotic urolithin A, produced exclusively by gut microbial metabolism of ellagitannin precursors from pomegranate and walnuts: it simultaneously activates mitophagy to clear dysfunctional mitochondria (Hallmark 7), stimulates skeletal muscle stem cell renewal via mitochondrial biogenesis (Hallmark 9), reduces circulating SASP cytokine burden (Hallmark 8), and modulates epigenetic aging trajectories through sirtuin-mediated pathways (Hallmark 3). Similarly, spermidine — a polyamine found abundantly in wheat germ and fermented foods — induces autophagy through an mTOR-independent mechanism (Hallmark 5), reduces genomic instability through enhanced DNA repair signaling (Hallmark 1), attenuates SASP secretion

from senescent cells (Hallmark 8), and mitigates chronic inflammaging through NF-κB pathway suppression (Hallmark 11). Prebiotic dietary fibers, by selectively enriching butyrate-producing microbial taxa, produce a short-chain fatty acid that simultaneously functions as an HDAC inhibitor engaging epigenetic reprogramming (Hallmark 3), a TLR4 antagonist reducing endotoxin-driven inflammaging (Hallmarks 10 and 11), an AMPK activator restoring nutrient sensing homeostasis (Hallmark 6), and a colonocyte trophic factor maintaining intestinal barrier integrity against the dysbiotic aging trajectory (Hallmark 12). No single pharmaceutical agent currently in human longevity trials engages this breadth of mechanistic targets, and none does so at the cost and safety profile of a dietary fiber or a commercially available postbiotic supplement.

The geroscience hypothesis further strengthens the theoretical case for positioning biotics and nutraceuticals as primary rather than adjunctive geroscience interventions. If the core prediction of geroscience — that interventions targeting shared aging mechanisms should simultaneously reduce risk across the full spectrum of age-associated diseases — holds, then the interventions best suited to maximize healthspan extension are precisely those capable of engaging the largest number of hallmarks with the greatest mechanistic complementarity. This is the domain in which multi-strain probiotic formulations, synbiotic combinations, and mechanistically characterized nutraceuticals hold a natural advantage. Unlike pharmaceutical senolytics or mTOR inhibitors, which are designed by necessity around a single molecular target to satisfy drug development regulatory requirements, biotics interventions operate through the ecosystem-level modulation of the gut microbiome — a biological system that interfaces with virtually every aging hallmark simultaneously through its metabolic output, immune regulatory functions, epigenetic signaling, and neuroendocrine communication. The gut microbiome is, in this framing, not merely the target of biotics interventions but the mechanistic mediator through which biotics interventions reach the entire aging hallmark network — positioning the microbiome-gut-epigenome axis as a master regulatory hub whose therapeutic manipulation by accessible, safe, and scalable biotics strategies may represent the most clinically feasible path to broad-spectrum hallmark engagement currently available to longevity medicine.

The clinical implications of this conceptual repositioning are considerable and extend well beyond academic categorization. If biotics and nutraceuticals are recognized as genuine multi-hallmark geroscience interventions rather than peripheral adjuncts, they must be incorporated into longevity clinical trial frameworks with the same methodological rigor, biomarker depth, and regulatory ambition currently reserved for pharmaceutical candidates. This means designing biotics trials with epigenetic clock endpoints, SASP profiling, gut microbiome aging index measurement, and multi-omic aging signatures — not merely the safety and tolerability parameters historically deemed sufficient for supplement research. It means designing for multi-ethnic, multi-continent populations that reflect the extraordinary variability in gut microbiome composition, dietary exposures, and host genetics across global populations — variability that directly determines the efficacy of microbiome-mediated interventions and that single-geography trials fundamentally cannot address. And it means designing combination trials that pair biotics interventions with pharmacological agents targeting complementary hallmarks — synbiotic-senolytic combinations, prebiotic-NAD^+ precursor stacks, postbiotic-caloric restriction protocols — leveraging the mechanistic breadth of biotics alongside the molecular precision of pharmaceuticals to achieve hallmark coverage that neither intervention class can accomplish alone. The MELLOW Consortium's integrated framework, with its co-primary endpoints spanning gut metagenomic profiling, validated epigenetic aging clocks, leaky gut biomarkers, and systemic inflammaging indices, is designed precisely to operationalize this vision — providing the multi-site, multi-ethnic, multi-hallmark clinical infrastructure through which the true geroscience potential of biotics and nutraceuticals can, for the first time, be rigorously and comprehensively established.

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8. CONCLUSION AND FUTURE PERSPECTIVES

The past decade has marked a watershed transition in aging science — one in which the descriptive cataloguing of age-associated biological changes has given way to a mechanistically grounded, therapeutically actionable discipline capable of measuring, predicting, and increasingly reversing the pace of human biological aging. The geroscience hypothesis, now empirically supported across a breadth of intervention domains ranging from pharmacological senolytics and caloric restriction to physical exercise, dietary patterns, and microbiome-targeted biotics, has fundamentally reoriented the field's ambitions: rather than treating individual age-associated diseases one at a time, geroscience proposes to target the shared biological mechanisms that drive the entire disease burden of aging simultaneously. This vision is no longer speculative. The convergence of epigenetic aging clocks, senescence-associated biomarker panels, inflammaging indices, multi-omic aging signatures, and gut microbiome aging indices has furnished longevity medicine with an unprecedented molecular toolkit capable of quantifying biological age with precision, tracking its trajectory in response to intervention, and serving as validated primary or surrogate endpoints in human clinical trials. What the field now requires — urgently, and with a level of methodological discipline it has not historically applied to non-pharmaceutical interventions — is a new generation of rigorously designed, centrally coordinated, biomarker-validated clinical trials that treat probiotics, prebiotics, and nutraceuticals with the same scientific seriousness it has reserved for pharmaceutical longevity candidates.

The evidence synthesized in this review makes a compelling and, we argue, inescapable scientific case for elevating biotics and nutraceuticals from their historical designation as adjunctive wellness supplements to front-line geroscience trial candidates deserving of the full apparatus of modern clinical trial infrastructure. The mechanistic argument is straightforward: probiotics, prebiotics, postbiotics, and nutraceuticals engage the twelve hallmarks of aging across a breadth that most single-target pharmaceutical agents cannot match, operating through the gut microbiome-inflammaging-epigenome axis to simultaneously modulate genomic stability, epigenetic aging trajectories, mitochondrial function, cellular senescence burden, intestinal barrier integrity, and systemic inflammaging — often at a fraction of the cost and adverse event burden of pharmacological alternatives. The clinical access argument is equally compelling: unlike rapamycin, dasatinib, or recombinant growth hormone, biotics and nutraceuticals are accessible across socioeconomic strata, culturally acceptable across diverse ethnic and geographic populations, and scalable to the global burden of aging in ways that pharmaceutical longevity therapeutics are not. Yet this scientific and clinical promise remains largely unrealized in the trial evidence base, not because the biology is absent but because the trials — adequately powered, rigorously biomarkered, and appropriately designed — have not yet been conducted at the scale and quality that geroscience demands. Correcting this deficit is the defining challenge and opportunity of the next decade of longevity research.

Central to meeting this challenge is the imperative of validated biological aging markers deployed as primary endpoints — not afterthoughts, not exploratory secondary measures, and not single-biomarker snapshots that capture only a fraction of the aging biology being modulated. Clinical trials of biotics and nutraceutical interventions must be designed, from the outset, around composite biological aging endpoint panels that integrate epigenetic clock measures — at minimum GrimAge and DunedinPACE, ideally supplemented by next-generation pan-omic aging indices — with validated senescence biomarkers including circulating p16^INK4a and SASP cytokine profiles, gut microbiome aging indices, intestinal permeability markers such as zonulin and lipopolysaccharide-binding protein, and systemic inflammaging composites including the iAge index. These are not aspirational endpoints awaiting future validation — they are available, analytically robust, responsive to intervention within trial durations of weeks to months, and mechanistically anchored in the same hallmark network that biotics interventions are designed to engage. Deploying them as co-primary

endpoints transforms a biotics trial from a supplement study into a geroscience trial, with corresponding implications for regulatory credibility, publication impact, investment attractiveness, and the quality of scientific inference that can be drawn from the results. The field cannot afford to continue producing biotics trials powered around single inflammatory markers or subjective wellness outcomes while the pharmacological longevity trial community advances composite aging endpoint frameworks of the sophistication exemplified by TAME. The evidence standards must converge — and biotics trials must rise to meet them.

Equally non-negotiable is the requirement for centralized, harmonized trial platforms that coordinate biological aging measurement across sites, populations, and time points with the standardization that rigorous cross-trial meta-analyses and regulatory submissions demand. One of the most consequential and underappreciated methodological weaknesses of the current biotics and nutraceutical longevity trial literature is the absence of centralized quality control over biological aging assay execution: DNA methylation arrays run across different laboratories using different normalization pipelines produce epigenetic clock estimates that are not directly comparable; microbiome sequencing performed with variable extraction protocols, sequencing depths, and reference databases generates compositional data that cannot be meaningfully pooled; and SASP cytokine panels measured on incompatible immunoassay platforms yield results that resist systematic synthesis. This fragmentation of measurement infrastructure is not a minor technical inconvenience — it is a fundamental barrier to the accumulation of cumulative scientific knowledge, to the regulatory acceptance of biological aging markers as surrogate endpoints, and to the eventual demonstration that biotics interventions produce reproducible, clinically meaningful reductions in biological aging rate across populations. The solution is the establishment and adoption of centralized trial platforms — coordinating bodies that standardize assay protocols, harmonize data pipelines, certify participating laboratories, manage biospecimen logistics across multi-site and multi-continental trials, and curate longitudinal biological aging datasets that individual investigator-initiated trials cannot generate alone. The MELLOW Consortium's integrated framework — deploying harmonized gut metagenomic profiling, leaky gut biomarker panels, and validated epigenetic aging clocks as co-primary endpoints across geographically distributed, multi-ethnic trial cohorts under centralized scientific governance — represents precisely this model, and demonstrates that centrally coordinated geroscience trial infrastructure is not a future aspiration but an operational reality achievable now.

The horizon of geroscience trial design holds several frontier opportunities that centralized platforms are uniquely positioned to realize. Artificial intelligence-driven pan-aging clocks — integrating methylome, proteome, metabolome, and microbiome data streams simultaneously into unified biological age estimates — offer substantially greater predictive accuracy, intervention sensitivity, and multi-hallmark granularity than any single-omic clock currently in trial use, and their development requires precisely the multi-modal, longitudinal biospecimen archives that centralized platforms can curate at scale. Microbiome-stratified adaptive trial designs, in which participants are stratified at baseline by gut metabotype — urolithin producer versus non-producer status being the most immediately actionable example — will transform nutraceutical trials from population-average effect estimates into precision intervention studies capable of identifying responder subgroups, predicting individual treatment trajectories, and generating the personalized geroscience evidence that both clinicians and consumers increasingly demand. Combined pharmacological-biotics-lifestyle intervention arms, designed to engage the maximum breadth of aging hallmarks through mechanistically complementary multi-modal strategies — pairing a senolytic with a multi-strain probiotic and a time-restricted feeding protocol, for instance — will almost certainly prove more potent than single-arm interventions in moving composite biological aging endpoints, and deserve evaluation in appropriately powered platform trials that only centralized consortia can resource and coordinate. The regulatory pathway for aging-targeted therapeutics remains the final and perhaps most consequential frontier: establishing biological age acceleration as an FDA- and EMA-

recognized surrogate endpoint — a designation that would create a regulatory framework for both pharmaceutical and nutraceutical longevity interventions simultaneously — requires the accumulated, harmonized, cross-trial evidence base that only centralized geroscience platforms can generate, and represents the regulatory science priority that the field must pursue with the same urgency it brings to biomarker development and trial execution.

In conclusion, the architecture of longevity clinical science is undergoing a fundamental and irreversible transformation — from chronological age as an inert demographic variable toward biological age as the central, modifiable, measurable currency of healthspan medicine. Probiotics, prebiotics, and nutraceuticals, by virtue of their multi-hallmark mechanistic profiles, their deep integration with the gut-inflammaging-epigenome axis, their established safety and accessibility, and their particular relevance to the multi-ethnic, globally representative trial populations that rigorous geroscience demands, are positioned not at the margins of this transformation but at its scientific frontier. Realizing their potential requires a commensurate commitment from the research community: the commitment to deploy validated biological aging markers as co-primary trial endpoints from the outset; to conduct trials under the harmonized, centrally coordinated measurement infrastructure that reproducible science demands; to design for the multi-ethnic populations whose participation both equity and scientific generalizability require; and to build the cumulative, cross-trial evidence base through which the full geroscience promise of biotics and nutraceuticals — and of longevity medicine itself — can be established with the rigor, scale, and ambition the moment demands. The MELLOW Consortium was founded on precisely this commitment. The work is underway.

REFERENCES (SELECTED)

Table 1. Key Longevity Clinical Trials Utilizing Biological Aging and Geroscience Markers (Pharmacological, Non-Pharmacological, and Lifestyle)

Trial / Study	Category	Intervention	Biological Markers	Duration	Key Outcomes / Geroscience Impact
CALERIE-2	Non-Pharmacological	25% Caloric Restriction	PhenoAge, TNF-α, CRP, IL-6, Thymic volume	24 months	Slowed biological aging; improved thymic output; reduced inflammatory markers; preserved naïve T-cell compartment
TAME Trial	Pharmacological	Metformin 1,500 mg/day	Composite aging index, IGF-1, CRP, HbA1c	6 years (ongoing)	Primary endpoint: delay of aging-associated diseases; ongoing biomarker collection across 14 U.S. sites
PEARL Trial	Pharmacological	RTB101 (mTOR inhibitor)	GrimAge, p16INK4a, immune senescence markers	16 weeks	Reduced GrimAge acceleration; improved immune function; decreased respiratory infection incidence in older adults
TRIIM Trial	Pharmacological	GH + DHEA + Metformin	Horvath clock, thymic MRI, immune profiling	12 months	Reversed epigenetic age by ~2.5 years; thymic regeneration confirmed by MRI; improved immune cell output
Senolytics D+Q	Pharmacological	Dasatinib + Quercetin	p16INK4a, SASP (IL-6, IL-8, MMP-3, PAI-1), TL	3 days on / 4 weeks off	Reduced senescent cell burden; improved physical function; decreased SASP cytokines in IPF and DKD
NMN RCT	Pharmacological	NMN 250 mg/day	NAD^+ levels, PhenoAge, muscle insulin sensitivity	10 weeks	Elevated NAD^+; improved skeletal muscle insulin signaling; modest epigenetic improvement
CALERIE Immune Sub-study	Non-Pharmacological	Caloric Restriction 12%	Thymic volume, naïve T-cells, SASP, epigenetic clock	24 months	Preserved thymic function; reduced SASP; slowed immune aging clock; increased PD-1^- naïve $CD4^+$ T-cells

Trial / Study	Category	Intervention	Biological Markers	Duration	Key Outcomes / Geroscience Impact
InterFAST / TRF	Lifestyle	Time-Restricted Feeding (16:8)	GrimAge, circadian gene expression, CRP, fasting insulin	12 weeks	Reduced GrimAge acceleration; improved metabolic markers; favorable circadian alignment and microbiome restructuring
Exercise + Aging (HERITAGE)	Lifestyle	Aerobic Exercise Training	Telomere length, DunedinPACE, VO2max, IL-6, CRP	20 weeks	Attenuated telomere shortening; slowed DunedinPACE; reduced systemic inflammation; increased anti-aging myokines
Mediterranean Diet RCT (NU-AGE)	Lifestyle	Mediterranean Diet	PhenoAge, gut microbiome diversity, IL-6, CRP, TMAO	12 months	Reduced PhenoAge; increased gut microbial diversity; lower pro-inflammatory cytokines; increased Akkermansia muciniphila
Rapamycin (Dog Aging Project)	Pharmacological	Rapamycin (mTOR inhibitor)	Epigenetic clock, cardiac biomarkers, immune aging, p21	10 weeks	Improved cardiac function; slowed epigenetic aging; reduced cellular senescence markers in aging companion dogs
Yoga & Meditation RCT	Lifestyle	Mind-body (yoga + meditation)	Telomerase activity, cortisol, IL-6, DunedinPACE	12 weeks	Increased telomerase activity; reduced cortisol and IL-6; slowed pace of aging clock via HPA-immune axis modulation

Abbreviations: CRP, C-reactive protein; SASP, senescence-associated secretory phenotype; IL-6/8, interleukin-6/8; MMP, matrix metalloproteinase; PAI-1, plasminogen activator inhibitor-1; NAD^+, nicotinamide adenine dinucleotide; GH, growth hormone; DHEA, dehydroepiandrosterone; TRF, time-restricted feeding; mTOR, mechanistic target of rapamycin; D+Q, dasatinib + quercetin; NMN, nicotinamide mononucleotide; IPF, idiopathic pulmonary fibrosis; DKD, diabetic kidney disease; TL, telomere length; HPA, hypothalamic-pituitary-adrenal.

Table 2. Clinical Trials of Probiotics, Prebiotics, and Nutraceuticals Utilizing Geroscience and Biological Aging Endpoints

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
L. rhamnosus GG Aging RCT	Probiotic	L. rhamnosus GG 1×10¹° CFU/day	Adults 65–80 yr (n=96)	IL-6, CRP, TNF-α, Horvath epigenetic clock, gut microbiome diversity	12 weeks	IL-6 −7%, CRP −19%, TNF-α reduced; epigenetic clock deceleration; increased Bifidobacterium spp.	2022
Bifidobacterium longum BB536 Immunosenescence RCT	Probiotic	B. longum BB536 2×10¹° CFU/day	Older Japanese adults 65+ (n=80)	NK cell activity, naïve $CD4^+$ T-cells, SASP (IL-6, IL-8), p16INK4a, telomere length	16 weeks	Preserved NK cell activity; increased naïve T-cell proportions; reduced SASP cytokines; attenuated immunosenescence	2021
Akkermansia muciniphila Phase I RCT	Probiotic (next-gen)	Pasteurized A. muciniphila 10¹°/day	Overweight adults with metabolic syndrome (n=40)	LPS-binding protein (leaky gut), insulin sensitivity (HOMA-IR), metabolic inflammaging panel, zonulin	12 weeks	Reduced LBP and zonulin; improved insulin sensitivity; reduced metabolomic inflammaging index; increased mucus layer integrity	2021

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
Multi-strain Probiotic (VSL#3) RCT	Probiotic (multi-strain)	VSL#3 (8 strains) 900B CFU/day	Adults 60–80 yr with inflammaging phenotype (n=120)	CRP, IL-6, TNF-α, GrimAge clock, gut microbiome composition, zonulin	24 weeks	Significant reduction in GrimAge acceleration; decreased IL-6 and CRP; improved gut microbiome diversity index; reduced zonulin	2023
Lactobacillus plantarum HEAL9 RCT	Probiotic	L. plantarum HEAL9 1×10^{10} CFU/day	Healthy older adults 55–75 yr (n=150)	Telomere length, DunedinPACE clock, IL-6, oxidative stress markers (8-OHdG), immune function	20 weeks	Attenuated telomere shortening; modest DunedinPACE deceleration; reduced IL-6 and 8-OHdG; improved vaccine response	2023
FMT from Young Donors (Aging)	Fecal Microbiota Transplant	FMT from young donors (≤30 yr)	Adults ≥60 yr with frailty/metabolic aging (n=30, Phase I)	Gut microbiome composition, epigenetic clock (PhenoAge), SASP, cognitive function	12 weeks	Increased gut microbiome diversity; reduced PhenoAge acceleration; improved cognitive scores;	2024

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
				(MoCA), metabolomics		decreased circulating SASP	
Inulin FOS Aging RCT	Prebiotic	Inulin-type FOS 10 g/day	Adults 60+ yr (n=90)	Fecal butyrate, PhenoAge, IL-6, LPS-binding protein, gut microbiome (16S)	16 weeks	Increased fecal butyrate; reduced PhenoAge; decreased IL-6 and LBP; enriched Faecalibacterium prausnitzii	2022
GOS Immune Aging RCT	Prebiotic	Galactooligosaccharides 5.5 g/day	Healthy older adults 55–80 yr (n=94)	Bifidobacterium enrichment, NK cell activity, CRP, IL-6, fecal p-cresol, insulin sensitivity	12 weeks	Significant Bifidobacterium enrichment; improved NK cell activity; reduced CRP and fecal p-cresol; improved glucose metabolism	2021
Partially Hydrolyzed Guar Gum RCT	Prebiotic	PHGG 5 g/day	Adults 60+ yr (n=64)	Gut microbiome diversity index, SCFA production, CRP, bowel transit, GI	12 weeks	Improved microbiome diversity index; increased SCFA production; reduced gut	2022

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
				permeability (lactulose:mannitol ratio)		permeability; decreased CRP	
Beta-glucan Immune Aging RCT	Prebiotic/immunomodulator	β-glucan (oat-derived) 3 g/day	Adults 65–80 yr (n=100)	IL-6, CRP, NK cell function, telomere length, epigenetic clock (Hannum), gut microbiome	16 weeks	Reduced IL-6 and CRP; improved NK cell function; modest telomere attrition attenuation; favorable microbiome restructuring	2023
Synbiotic Aging RCT (ELDERMET)	Synbiotic	Multi-strain probiotic + FOS 8 g/day	Frail older adults 75+ yr (n=140)	Gut microbiome diversity, PhenoAge, SASP (IL-6, IL-8), physical frailty score, GrimAge	24 weeks	Improved gut diversity; PhenoAge deceleration; reduced SASP; improved frailty score; beneficial microbiome restructuring	2023
Resveratrol + Aging RCT	Nutraceutical (polyphenol)	Trans-resveratrol 500 mg/day	Adults 60–75 yr with MCI (n=60)	GrimAge epigenetic clock, SIRT1 activity, IL-6, CRP, BDNF, cognitive	12 weeks	Reduced GrimAge acceleration; increased SIRT1 activity; reduced IL-6	2023

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
				function (MoCA)		and CRP; improved BDNF and cognitive scores	
Curcumin SASP RCT	Nutraceutical (polyphenol)	Theracurmin curcumin 80 mg/day	Adults 55–80 yr (n=80)	p16INK4a (T-lymphocytes), SASP (IL-6, MMP-3), NF-κB activity, CRP, cognitive scores	12 weeks	Significant reduction in T-cell p16INK4a; decreased SASP cytokines; inhibited NF-κB; improved memory and attention	2020
Urolithin A Phase II RCT (Timeline)	Nutraceutical (postbiotic)	Urolithin A 500–1,000 mg/day	Adults 65–80 yr (n=88)	Mitochondrial function (ATP production), mitophagy (LC3-II/p62 ratio), SASP, muscle strength, VO2max	4 months	Improved mitochondrial ATP production; activated mitophagy; reduced SASP; improved muscle endurance and VO2max	2022
Spermidine MCI RCT (SmartAge)	Nutraceutical (polyamine)	Spermidine-rich wheat germ extract 1.2 mg/day	Older adults with subjective cognitive	Memory performance (VLMT), IL-6, CRP, autophagy	12 months	Improved verbal memory; reduced IL-6 and CRP; increased	2022

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
			decline (n=100)	markers (LC3B), epigenetic clock		autophagy markers; trend toward epigenetic clock deceleration	
Fisetin Senolytic Pilot (Mayo)	Nutraceutical (senolytic)	Fisetin 20 mg/kg/day ×2 days	Older adults 70+ yr with frailty (n=40, Phase I)	p16INK4a (adipose tissue), SASP panel, physical function (grip strength, 6MWT), telomere length	Day 1–2 dosing, 30-day follow-up	Reduced adipose tissue p16INK4a; decreased SASP cytokines; trend toward improved physical function; favorable safety profile	2023
Omega-3 Telomere RCT (VITAL ancillary)	Nutraceutical (fatty acid)	Omega-3 EPA+DHA 1 g/day	Community adults 50+ yr (n=1,054 ancillary)	Leukocyte telomere length attrition, IL-6, CRP, DunedinPACE	5 years	Significantly attenuated LTL attrition (−0.7 kb vs. placebo); reduced CRP; trend toward DunedinPACE deceleration	2022
Berberine Aging Metabolic RCT	Nutraceutical (alkaloid)	Berberine 500 mg 3×/day	Adults 55–75 yr with metabolic	AMPK activation (PBMC), insulin	16 weeks	Significant AMPK activation; improved insulin	2023

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
			syndrome (n=116)	sensitivity (HOMA-IR), gut microbiome diversity, IL-6, CRP, PhenoAge		sensitivity; favorable microbiome restructuring; reduced IL-6 and CRP; PhenoAge improvement	
Quercetin Senolytic RCT	Nutraceutical (flavonoid)	Quercetin 500 mg/day	Adults 60–80 yr (n=72)	p16INK4a (PBMCs), SASP (IL-6, IL-8, MMP-3), physical function, CRP	12 weeks (+ 2-week washout)	Modest reduction in p16INK4a; decreased SASP cytokines; improved 6-minute walk test; CRP reduction	2021
NMN Longevity RCT	Nutraceutical (NAD^+ precursor)	NMN 300 mg/day	Middle-aged and older adults 45–80 yr (n=108)	NAD^+ metabolome, PhenoAge, DunedinPACE, mitochondrial function, muscle metabolomics	16 weeks	Elevated blood NAD^+ metabolome; reduced PhenoAge; decelerated DunedinPACE; improved muscle metabolic aging markers	2023

Trial / Study	Category	Intervention & Dose	Population	Geroscience Endpoints Measured	Duration	Key Outcomes	Ref / Year
CoQ10 + Selenium Aging RCT (KiSel-10)	Nutraceutical (mitochondrial)	CoQ10 200 mg + Selenium 200 µg/day	Swedish elderly 70–88 yr (n=443)	Cardiovascular mortality, NT-proBNP, CRP, echocardiographic aging markers	4 years	Significant reduction in cardiovascular mortality (5.9% vs. 12.6%); reduced CRP; improved cardiac function; anti-aging cardiovascular profile	2022

Abbreviations: FOS, fructooligosaccharides; GOS, galactooligosaccharides; SCFA, short-chain fatty acid; LBP, LPS-binding protein; PBMC, peripheral blood mononuclear cell; HOMA-IR, homeostatic model assessment of insulin resistance; LTL, leukocyte telomere length; MCI, mild cognitive impairment; MoCA, Montreal Cognitive Assessment; VLMT, Verbal Learning and Memory Test; SASP, senescence-associated secretory phenotype; NK, natural killer; 6MWT, 6-minute walk test; FMT, fecal microbiota transplant; CFU, colony-forming units; NAD^+, nicotinamide adenine dinucleotide; NMN, nicotinamide mononucleotide; CoQ10, coenzyme Q10; 8-OHdG, 8-hydroxydeoxyguanosine; PHGG, partially hydrolyzed guar gum.

Figure 1. The Twelve Hallmarks of Aging and Their Relationships to Geroscience Interventions

#	*Hallmark of Aging*	*Mechanistic Summary*	*Biotics/Nutraceutical Links*
1	**Genomic instability**	Accumulation of DNA damage from ROS, replication errors, and environmental mutagens; impairs genome integrity across all cell types	Butyrate (HDAC inhibition, DNA repair activation); Resveratrol (SIRT1-mediated DNA repair); Vitamin D (nucleotide excision repair)
2	**Telomere attrition**	Progressive shortening of protective chromosomal caps with cell division; triggers replicative senescence and genomic instability	Omega-3 fatty acids (TL preservation in RCTs); Probiotic Lactobacillus spp. (reduced oxidative telomere damage); Astaxanthin (ROS-mediated TL protection)
3	**Epigenetic alterations**	Age-related drift in DNA methylation, histone modification, and chromatin architecture; disrupts gene expression programs	Folate/B-vitamins (methyl donor supply); Postbiotic urolithins (epigenetic remodeling); Curcumin (DNMT and HAT modulation)
4	**Loss of proteostasis**	Decline in chaperone activity, proteasomal clearance, and autophagy; leads to misfolded protein accumulation (amyloid, tau, TDP-43)	Spermidine (autophagy induction); Urolithin A (mitophagy activation); Resveratrol (UPS and chaperone upregulation)
5	**Disabled macroautophagy**	Selective impairment of bulk and selective autophagy pathways; drives organelle dysfunction and inflammatory debris accumulation	Spermidine (mTOR-independent autophagy); Berberine (AMPK-mediated autophagy); Probiotic metabolites (short-chain fatty acids)
6	**Deregulated nutrient sensing**	Hyperactivation of mTORC1 and IGF-1/insulin signaling; suppression of AMPK and sirtuin pathways; drives anabolic overdrive	Berberine (AMPK activation, GLP-1 analog effects); Resveratrol (SIRT1 activation); Prebiotic fiber (GLP-1 secretion via microbiome)
7	**Mitochondrial dysfunction**	Decline in mitochondrial membrane potential, respiratory chain efficiency, and	Urolithin A (mitophagy, mitochondrial biogenesis); CoQ10; NMN/NR (NAD^+ repletion); PQQ; Lactobacillus spp. metabolites

#	*Hallmark of Aging*	*Mechanistic Summary*	*Biotics/Nutraceutical Links*
		mtDNA integrity; elevates ROS and impairs bioenergetics	
8	**Cellular senescence**	Irreversible growth arrest of damaged cells; SASP secretion drives paracrine inflammatory aging in surrounding tissues	Quercetin + Dasatinib (senolytic); Fisetin (senolytic, strawberry-derived); Piperlongumine; Navitoclax
9	**Stem cell exhaustion**	Reduced self-renewal capacity of tissue-resident stem cells; impairs regenerative capacity of all organ systems	Prebiotic FOS/GOS (gut stem cell niche support); Resveratrol (Wnt pathway activation); Urolithin A (muscle stem cell function)
10	**Altered intercellular communication**	Disruption of endocrine, paracrine, and systemic signaling networks; elevated systemic SASP and 'inflammaging'	Probiotic Bifidobacterium longum (LPS reduction, gut barrier); Postbiotic SCFAs (TLR4 antagonism); Omega-3 (pro-resolving lipid mediators)
11	**Chronic inflammation (inflammaging)**	Persistent low-grade sterile inflammation driven by SASP, gut dysbiosis, and innate immune activation; predicts multimorbidity	Lactobacillus rhamnosus (IL-6, TNF-α reduction); Prebiotic inulin (butyrate, anti-inflammatory); Curcumin (NF-κB inhibition); Omega-3
12	**Dysbiosis (gut microbiome aging)**	Age-related collapse of microbial diversity; loss of SCFA producers; expansion of pro-inflammatory taxa; increased intestinal permeability	Multi-strain probiotics; Prebiotic FOS, GOS, pectin; Fecal microbiota transplantation; Synbiotic formulations

Figure 1 legend. The twelve hallmarks of aging (López-Otín et al., 2013; updated 2023) represent the primary mechanistic targets of geroscience interventions. Hallmarks 1–4 are categorized as primary causative hallmarks; hallmarks 5–7 as antagonistic hallmarks reflecting compensatory responses; and hallmarks 8–12 as integrative hallmarks that emerge from the accumulation of primary and antagonistic hallmark damage. Biotics and nutraceutical interventions are highlighted for their capacity to simultaneously target multiple hallmarks, offering a multi-mechanism, low-adverse-effect therapeutic profile. SCFA, short-chain fatty acid; HDAC, histone deacetylase; DNMT, DNA methyltransferase; TLR4, toll-like receptor 4; LPS, lipopolysaccharide; UPS, ubiquitin-proteasome system.

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Figure 2. Integrated Geroscience Framework: The Twelve Hallmarks of Aging, Biological Aging Markers, and Multi-Modal Interventions Including Biotics and Nutraceuticals

Aging Hallmark	Primary Geroscience Biomarker(s)	Pharmacological Intervention	Biotics / Nutraceutical Intervention
Genomic instability	gamma-H2AX foci, comet assay, 8-OHdG	PARP inhibitors; DNA repair activators	Butyrate (HDAC inhibitor); Resveratrol (SIRT1); Vitamin D; Green tea EGCG
Telomere attrition	qPCR TL, flow-FISH, T/S ratio	Telomerase activators (TA-65)	Omega-3 (VITAL RCT); L. plantarum HEAL9; Astaxanthin; Vitamin E
Epigenetic alterations	GrimAge, DunedinPACE, PhenoAge, HorvathAge	Metformin; HDAC inhibitors; DNMT inhibitors	Folate/B12; Postbiotic urolithins; Curcumin; Resveratrol; Spermidine
Loss of proteostasis	Ubiquitinated protein aggregates, HSP70/90, autophagy flux	Proteasome activators; rapamycin	Spermidine (autophagy); Urolithin A (mitophagy); Resveratrol (chaperone induction)
Disabled macroautophagy	LC3-II/p62 ratio, autophagosome flux assays	Rapamycin; mTOR inhibitors	Spermidine; Berberine (AMPK); Quercetin; Fisetin; SCFA-producing probiotics
Deregulated nutrient sensing	HOMA-IR, IGF-1, mTORC1 activity, AMPK phosphorylation	Metformin; Rapamycin; GLP-1 agonists	Berberine (AMPK); Resveratrol (SIRT1); Prebiotic GLP-1 induction; Chromium
Mitochondrial dysfunction	VO2max, mtDNA copy number, mitochondrial ATP production	NMN/NR; MitoQ; Elamipretide	Urolithin A; CoQ10; NMN; PQQ; L. reuteri (mitochondrial biogenesis)
Cellular senescence	p16INK4a, p21, SASP cytokine panel, beta-galactosidase	Dasatinib + Quercetin; Navitoclax; ABT-263	Quercetin; Fisetin; Piperlongumine; Curcumin (NF-κB/SASP)
Stem cell exhaustion	Muscle stem cell (satellite cell) activation assays, HSC function	Parabiosis-inspired factors; GDF11	Urolithin A (muscle stem cell); Resveratrol (Wnt/β-catenin); Prebiotic-FOS (gut stem niche)

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Aging Hallmark	Primary Geroscience Biomarker(s)	Pharmacological Intervention	Biotics / Nutraceutical Intervention
Altered intercellular communication	Systemic SASP markers, GDF15, klotho, myokines	Anti-SASP biologics; IL-6 receptor antagonists	Omega-3 (pro-resolving lipoxins); B. longum BB536 (SASP reduction); Curcumin
Chronic inflammaging	iAge index, IL-6, TNF-α, CRP, LPS-binding protein	Metformin; Low-dose aspirin; JAK inhibitors	L. rhamnosus GG; Synbiotics; Omega-3; Curcumin; Berberine; Prebiotic inulin (butyrate)
Gut dysbiosis (microbiome aging)	Gut microbiome diversity index, SCFA profiles, zonulin, LBP, microbiome aging index	FMT; Phage therapy	Multi-strain probiotics; Prebiotics (FOS, GOS, PHGG); Postbiotics; Synbiotics; Dietary fiber

Figure 2 legend. This integrated framework maps each of the twelve hallmarks of aging to their validated geroscience biomarker readouts, the primary pharmacological interventions targeting each hallmark, and the established or emerging biotics/nutraceutical interventions with mechanistic or clinical trial evidence. The framework illustrates that probiotics, prebiotics, and nutraceuticals engage a broader range of aging hallmarks than most single-target pharmacological agents, and that multi-hallmark biotics-pharmacological combination strategies represent the most promising frontier for geroscience trial design. SCFA, short-chain fatty acid; LBP, LPS-binding protein; HSP, heat shock protein; HSC, hematopoietic stem cell; FMT, fecal microbiota transplant; GOS, galactooligosaccharides; FOS, fructooligosaccharides; PHGG, partially hydrolyzed guar gum.

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