Consciously Take Back Control of the Aging Process

Arland Hill, DC, MPH, DACBN

Aging: Is it Optional?

- Can we actually slow the aging process
- “My father didn’t live long, so I probably won’t either.”
- Genetic factors
- Non-genetic factors (epigenetics)
- Increased risk of many chronic diseases with aging
  - Neurodegenerative
  - Cerebrovascular
  - Cancer

Chronological vs. Biological Aging

- Lifespan vs. Healthspan
- Is lifespan a good metric?
  - Research interpretation
  - Practical observation
- It's not the days you live, but how you live those days.
- Hallmarks of aging only came about in 2013

www.nature.com/mp Molecular Psychiatry

EXPERT REVIEW OPEN

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Targeting the "hallmarks of aging" to slow aging and treat age-related disease: fact or fiction?

Maryam Keshavarz[1], Kan Xie[1], Kristina Schaaf[1], Daniele Bano[2] and Dan Ehninger[1]✉

Aging is a major risk factor for a number of chronic diseases, including neurodegenerative and cerebrovascular disorders. Aging processes have therefore been discussed as potential targets for the development of novel and broadly effective preventatives or therapeutics for age-related diseases, including those affecting the brain. Mechanisms thought to contribute to aging have been summarized under the term the "hallmarks of aging" and include a loss of proteostasis, mitochondrial dysfunction, altered nutrient sensing, telomere attrition, genomic instability, cellular senescence, stem cell exhaustion, epigenetic alterations and altered intercellular communication. We here examine key claims about the "hallmarks of aging". Our analysis reveals important weaknesses that preclude strong and definitive conclusions concerning a possible role of these processes in shaping organismal aging rate. Significant ambiguity arises from the overreliance on lifespan as a proxy marker for aging, the use of models with unclear relevance for organismal aging, and the use of study designs that do not allow to properly estimate intervention effects on aging rate. We also discuss future research directions that should be taken to clarify if and to what extent putative aging regulators do in fact interact with aging. These include multidimensional analytical frameworks as well as designs that facilitate the proper assessment of intervention effects on aging rate.

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INTRODUCTION

Life expectancy increased from ~50 years in the early 1900s to over 80 years at present [1]. Factors contributing to this development may include advances in medical care as well as the creation of cleaner, safer, and healthier environments for people to live in [1]. Although this represents a great achievement for human societies, the growth in both the size and the proportion of the elderly population also comes with critical challenges. Advanced age is the main risk factor for many common diseases, such as cancers, cardiovascular disorders, and neurodegeneration [1]. Age-related neurodegenerative diseases, including Alzheimer's disease (AD), Parkinson's disease (PD), amyotrophic lateral sclerosis (ALS), and others [2–4], severely compromise the quality of life of affected individuals. Moreover, current demographic developments have substantial socioeconomic implications for health and care systems [5, 6]. Available treatments are symptomatic, despite intensive efforts to develop disease-modifying therapies for these devastating conditions [5, 6].

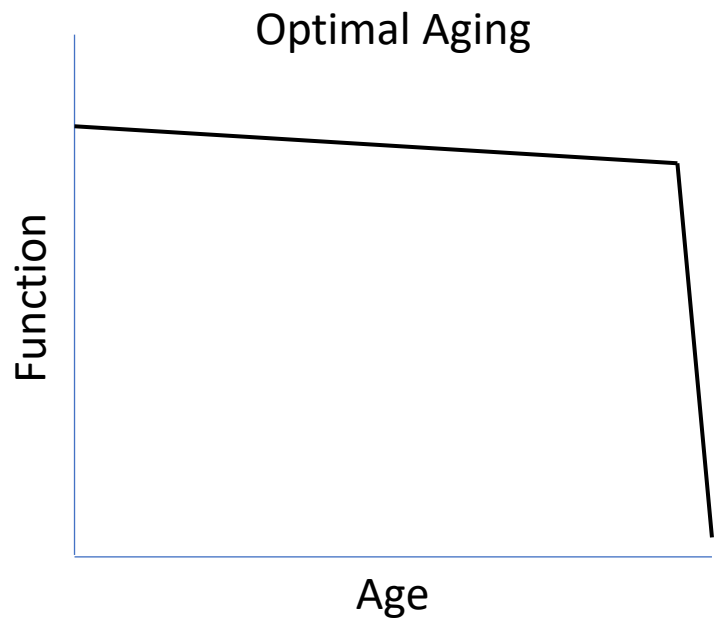
Among known risk factors for neurodegenerative diseases, the aging process itself has the highest impact [7]. For instance, it has been estimated that the risk for developing AD doubles every 5 years over the age of 65; the risk of death due to AD increases 700-fold between the ages 55 and 85 [8, 9]. Hence, strong mechanistic links between brain aging and neurodegenerative disease have been considered [10] and treatments with putative anti-aging drugs (e.g., rapamycin) have been proposed for clinical trials targeting AD [9]. Thus, studying aging and understanding how exactly aging increases the risk to develop neurodegenerative diseases can provide important clues to inform new strategies for early detection, prevention, and treatment.

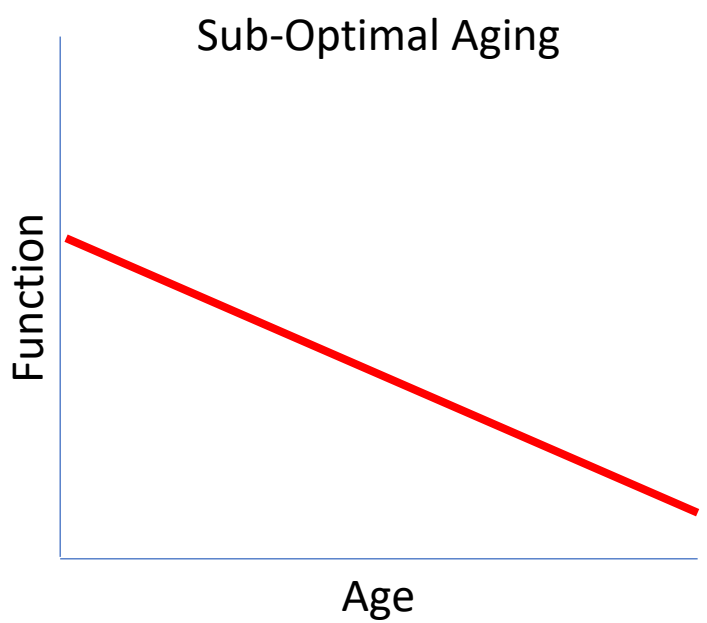
The critical outstanding question is: Can aging processes be slowed down? Evidence in nature suggests a positive answer to this fundamental question. For instance, similar pathobiological changes associated with aging develop over very different time scales in different mammalian species [11]. While it may take 70 years for a senile cataract to develop in a human, similar age-related changes develop in horses within 20 years, in dogs within 10 years, and in mice in even only 2 years. Analogous considerations also apply to many other age-related alterations (hair greying, muscle loss, etc.). Although the biology underlying these differences in aging rate are not well understood, these examples demonstrate that similar aging phenomena in comparable tissues can develop over very different absolute time scales. Therefore, there seems to be some plasticity that could be harnessed, in theory, for slowing down the aging process.

Much of what is currently thought to be known about the biological underpinnings of the aging process has been presented in concepts like the "hallmarks of aging" [12–14] which summarize processes claimed to be involved in driving organismal aging phenomena. Here, we carefully examine the evidence presented in favor of such links between these processes and aging. As we will explain in detail below, we identify limitations that are often grounded in the choice of models and/or the way aging is

Taking Back Control of the Aging Process

- Goal – Have a sharp and dramatic decline in function at the end of life
- Avoid slow, steady decline in function





Hallmarks of Aging Outline

- The 12 Hallmarks of Aging
- Identifying Inflammatory Sources and Their Link to Aging
- Gut-Aging Foundation
- Protein: Underuse and Impact
- Wake up Your Cells for Resiliency

12 Hallmarks of Aging

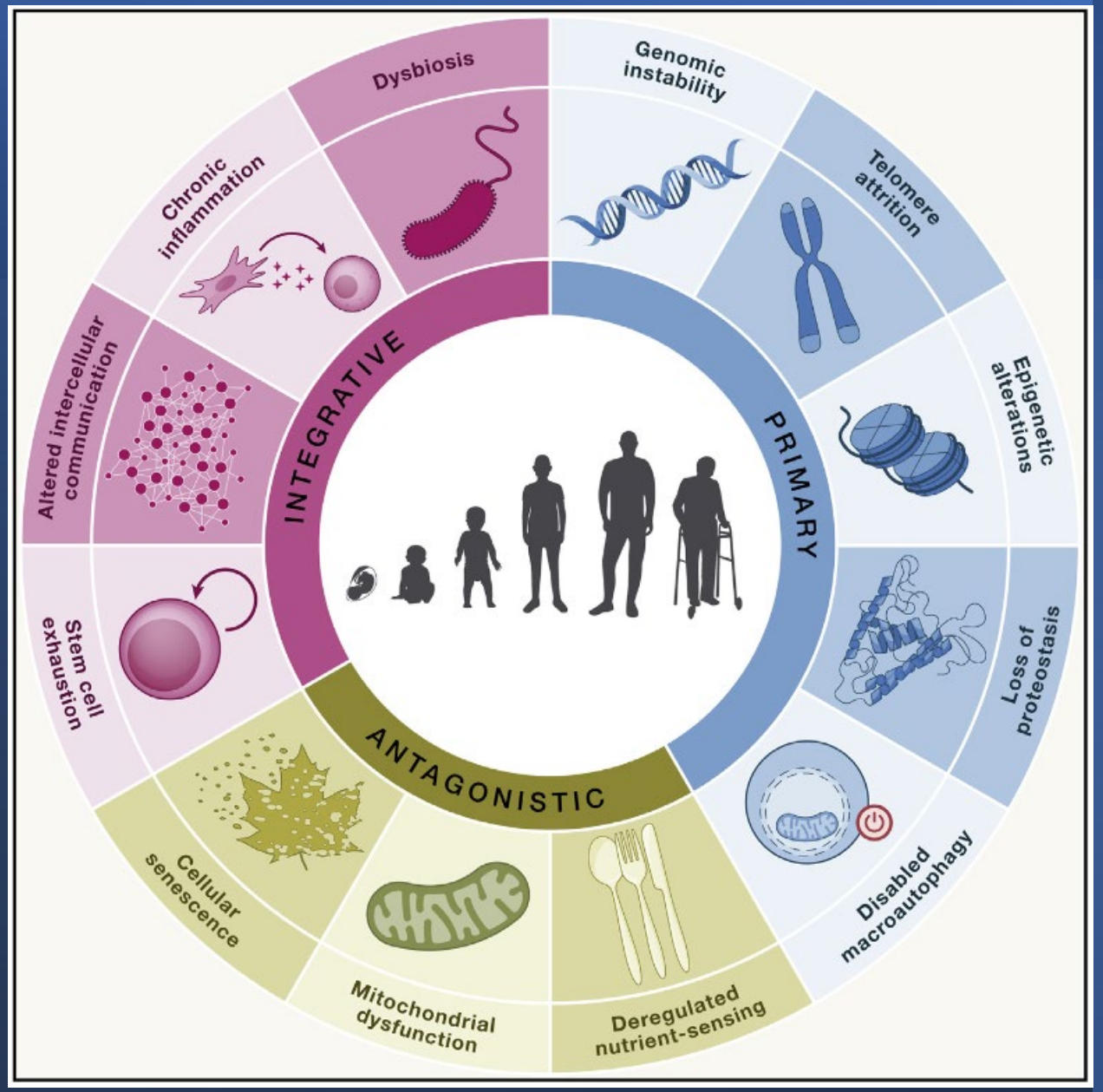
12 Interdependent Relationships

What is Aging

- The gradual decline in the physiological functions of an organism over time that is necessary for survival and reproduction.
- Physical changes of aging are not synonymous with diseases of aging.

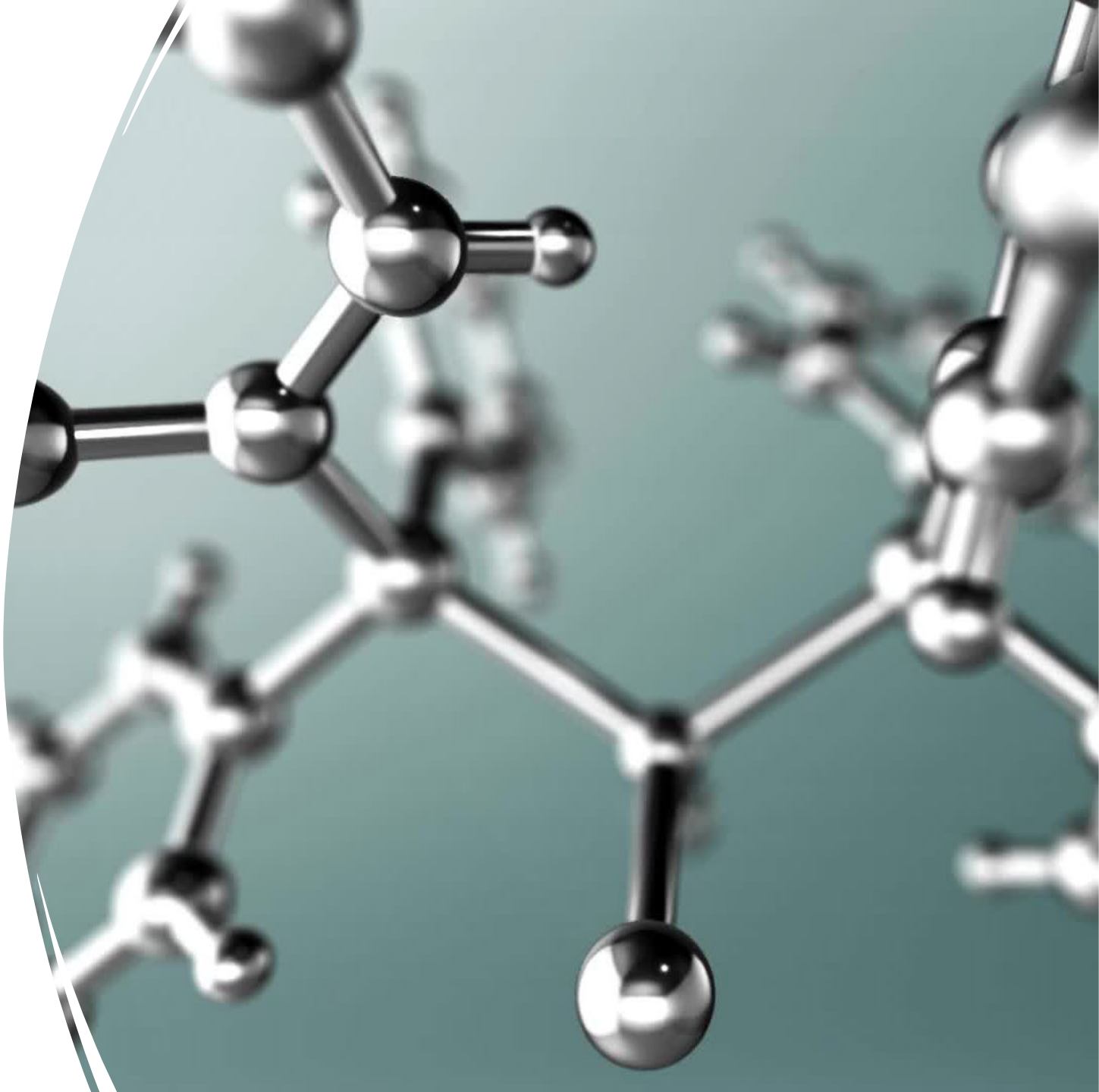
Aging Linked to Function

- Genomic instability
- Telomere attrition
- Epigenetic alterations
- Loss of proteostasis
- Disabled macroautophagy
- Deregulated nutrient-sensing
- Mitochondrial dysfunction
- Cellular senescence
- Stem cell exhaustion
- Altered intercellular communication
- Chronic inflammation
- Dysbiosis


Dysbiosis
Genomic instability
Telomere attrition
Epigenetic alterations
Loss of proteostasis
Disabled macroautophagy
Deregulated nutrient-sensing
Mitochondrial dysfunction
Cellular senescence
Stem cell exhaustion
Altered intercellular communication
Chronic inflammation
INTEGRATIVE
PRIMARY
ANTAGONISTIC

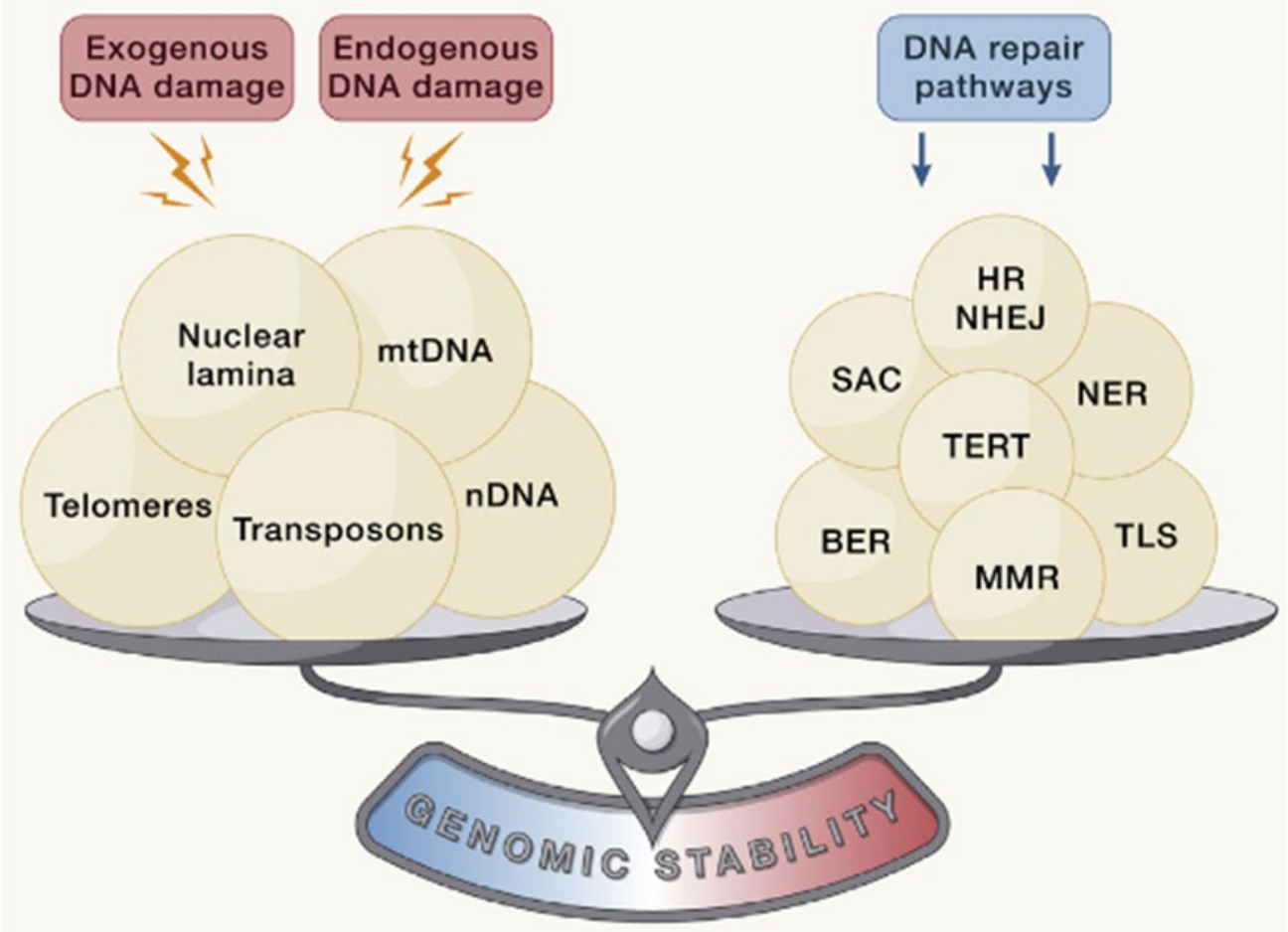
Genomic Instability

- External threats - physical, chemical or biological agents
- Internal threats – DNA replication errors, oxidative and hydrolysis reactions
- Gene disruptions
- Can be corrected by DNA repair and maintenance mechanisms
- These mechanisms lose efficacy over time



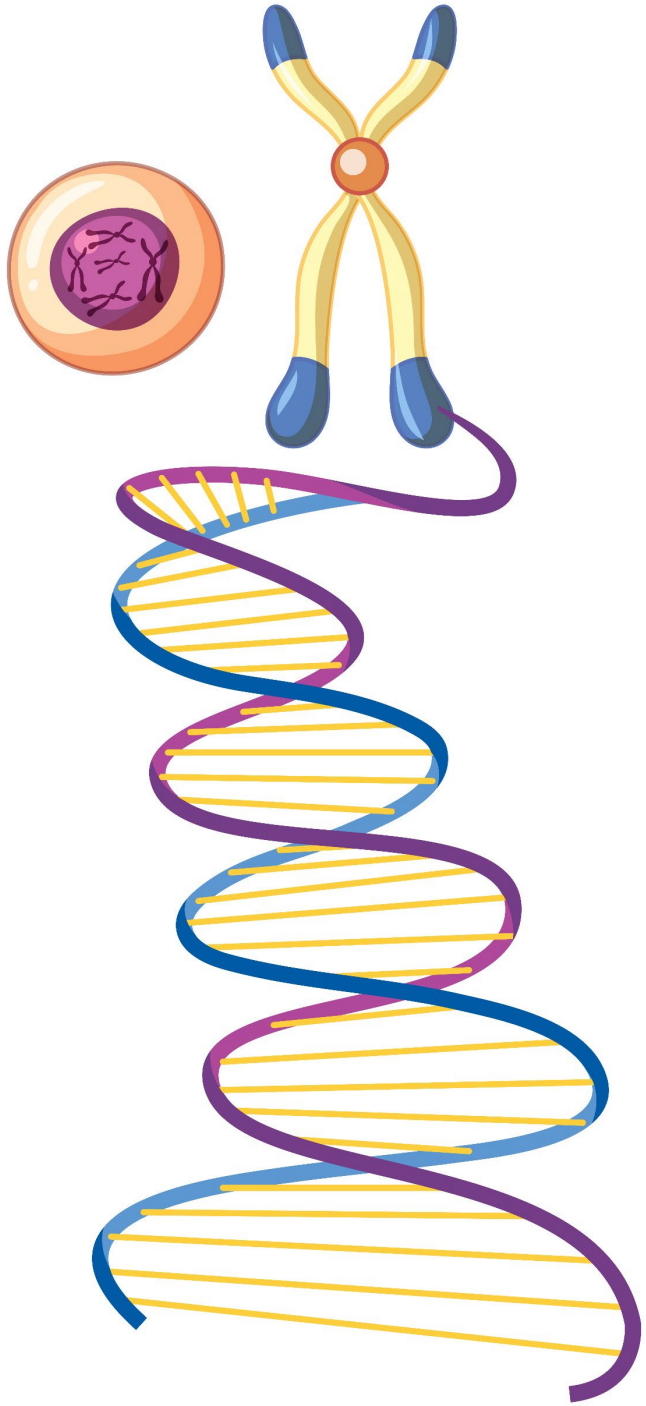
Genomic Instability

- Nuclear DNA damage
- Mitochondrial DNA damage
- DNA damage is persistent
- Balance between damage and repair
- Adequate repair – slower aging
- Inefficient repair – faster aging
- Think patient healing time



Telomere Attrition

- Telomere – end of the chromosomes
- Wearing down
- DNA damage to the end of the chromosomes
- Shorter – aging faster
- Longer – aging slower



Telomere Attrition

Conclusion: Telomere shortening is seen even at the stage of IGT (impaired glucose tolerance). Among subjects with Type 2 diabetes, those with atherosclerotic plaques had greater shortening of telomere length compared to those without plaques.

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Association of telomere shortening with impaired glucose tolerance and diabetic macroangiopathy

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Abstract

Objective: Shortening of telomere length has been reported in several conditions including Type 2 diabetes and atherosclerosis. The aims of this study were (1) to assess whether telomere shortening occurs at the stage of pre-diabetes, i.e., impaired glucose tolerance (IGT) and (2) whether telomere shortening was greater in Type 2 diabetic subjects with atherosclerotic plaques.
Methods: Subjects with impaired glucose tolerance (IGT) ($n=30$), non-diabetic control subjects ($n=30$), Type 2 diabetic patients without ($n=30$) and with atherosclerotic plaques ($n=30$) were selected from the Chennai Urban Rural Epidemiology Study (CURES), an ongoing epidemiological population-based study. Southern-blot analysis was used to determine mean terminal restriction fragment (TRF) length, a measure of average telomere size, in leukocyte DNA. Levels of thiobarbituric acid reactive substances (TBARS), protein carbonyl content (PCO) and high sensitive C-reactive protein (hs-CRP) were measured by standard methodologies. Carotid intima-media thickness (IMT) was assessed by high resolution B-mode ultrasonography.
Results: The mean (±S.E.) TRF lengths were significantly lower in IGT subjects (6.97 ± 0.3 kb; $p=0.002$) and lower still in Type 2 diabetic subjects without plaques (6.21 ± 0.2; $p=0.0001$) and lowest in Type 2 diabetic subjects with atherosclerotic plaques (5.39 ± 0.2; $p=0.0001$) when compared to control subjects (8.7 ± 0.5). In IGT subjects, TRF length was positively correlated to HDL cholesterol and negatively correlated to glycated hemoglobin (HbA_{1c}), TBARS, PCO, HOMA-IR and IMT. In multiple linear regression analysis, presence of diabetes, HDL cholesterol and increased TBARS levels appear as significant determinants of telomere shortening.
Conclusion: Telomere shortening is seen even at the stage of IGT. Among subjects with Type 2 diabetes, those with atherosclerotic plaques had greater shortening of telomere length compared to those without plaques.

Keywords: Oxidative stress; Telomere shortening; Macroangiopathy; IGT

Telomere Attrition

Telomere lengths in the white blood cells reflect skeletal muscle telomere length

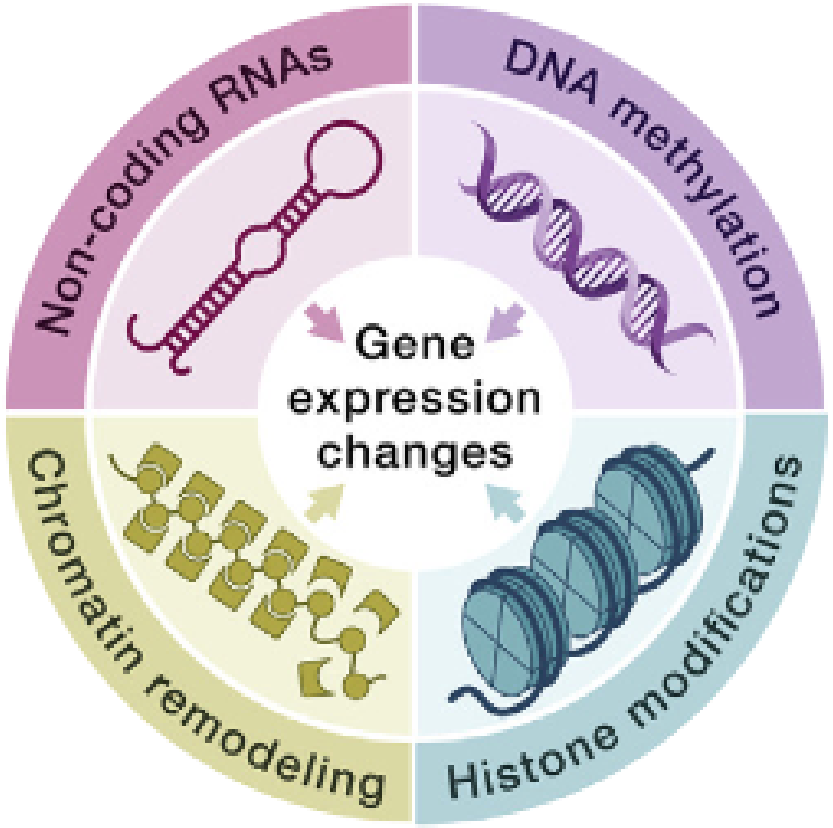
How does this affect your musculoskeletal patients?

Telomere Attrition

- Telomerase – enzyme that maintains telomere length
- Shortening is observed throughout life
- Activity preserved by nutrients such as magnesium
- Deficiency leads to disease

Epigenetic Alterations

- Environmental influences over gene expression
- DNA methylation
- Modification of histones
- Abberant chromatin remodeling
- Deregulated RNA function
- Influence over cancer, neurodegeneration, metabolic syndrome, and bone diseases



Loss of Proteostasis

Impaired protein homeostasis

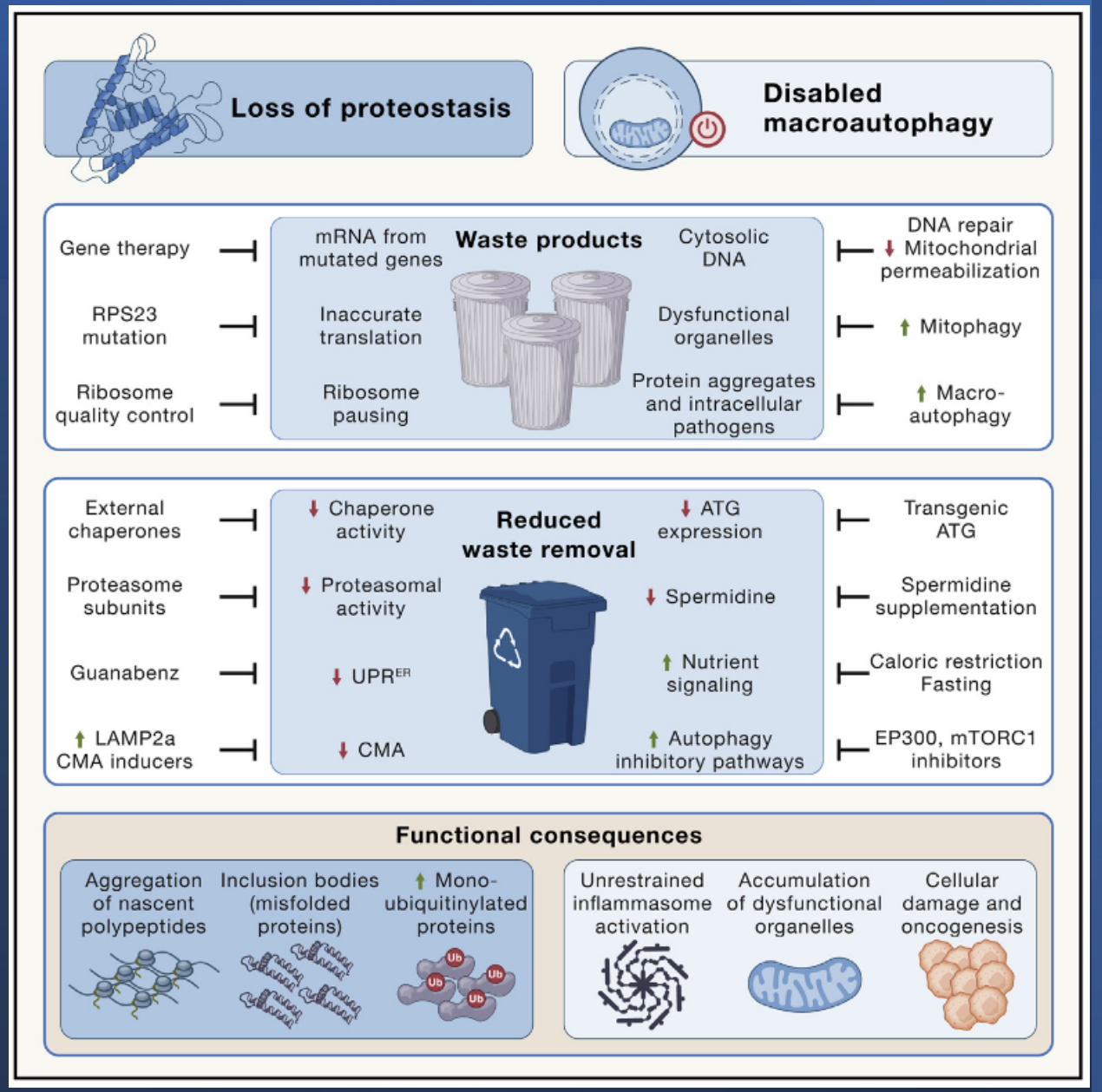
Leads to misfolded, oxidized, glycated or ubiquitinylated proteins

Forms aggregates that lead to plaques

Linked to multiple conditions

- Amyotrophic lateral sclerosis (ALS)
- Alzheimer’s Disease
- Parkinson’s Disease
- Cataracts

Balanced with autophagy

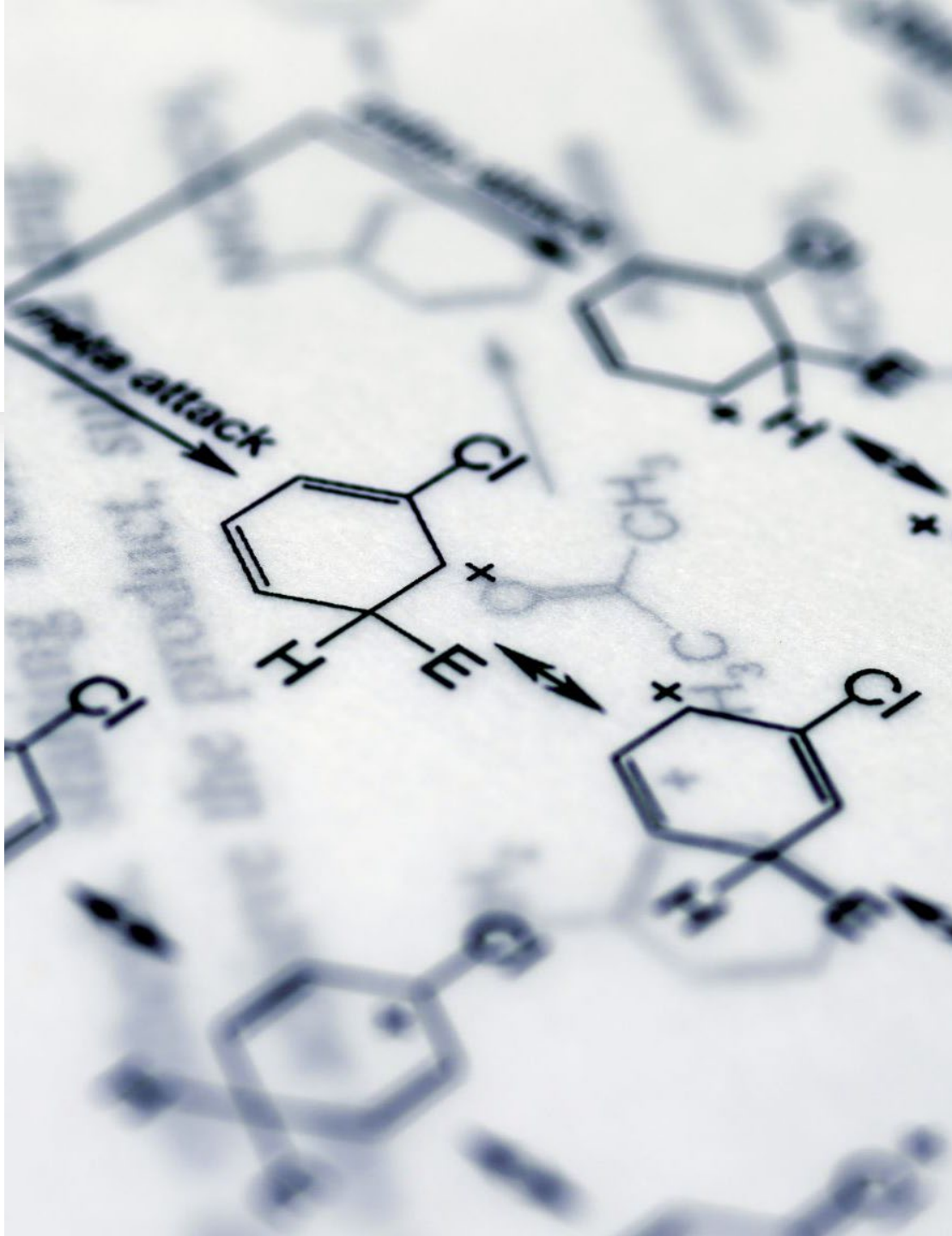

Loss of proteostasis
Disabled macroautophagy
Gene therapy
RPS23 mutation
Ribosome quality control
mRNA from mutated genes
Inaccurate translation
Ribosome pausing
Waste products
Cytosolic DNA
Dysfunctional organelles
Protein aggregates and intracellular pathogens
DNA repair
↓ Mitochondrial permeabilization
↑ Mitophagy
↑ Macro-autophagy
External chaperones
Proteasome subunits
Guanabenz
↑ LAMP2a CMA inducers
↓ Chaperone activity
↓ Proteasomal activity
↓ UPR^ER
↓ CMA
Reduced waste removal
↓ ATG expression
↓ Spermidine
↑ Nutrient signaling
↑ Autophagy inhibitory pathways
Transgenic ATG
Spermidine supplementation
Caloric restriction Fasting
EP300, mTORC1 inhibitors
Functional consequences
Aggregation of nascent polypeptides
Inclusion bodies (misfolded proteins)
↑ Mono-ubiquitinylated proteins
Ub
Unrestrained inflammasome activation
Accumulation of dysfunctional organelles
Cellular damage and oncogenesis

Disabled Macroautophagy (aka Autophagy)

- The opposite side of loss of proteostasis
- “Clean up process”
- Removing dysfunctional cellular material
- Sequestration of cellular material into vesicles that fuse with lysosomes for digestion / degradation
- Affects non-protein structures as well
- Ex. Removing dysfunction mitochondria (mitophagy)
- Ex. Invading pathogens (xenophagy)

Deregulated Nutrient-Sensing

- Insulin, IGF
- MTOR (mechanistic target of rapamycin)
  - Prolonged vs. short term stimulation
- Responds to:
  - Nutrients (glucose and amino acids)
  - Stressors (hypoxia, low energy)
- Regulates cellular activity
  - Autophagy
  - Biogenesis of mRNA, ribosomes and mitochondria
  - Protein synthesis
  - Glucose
  - Nucleotide and lipid metabolism



Deregulated Nutrient-Sensing

Activates anabolism

- Nutrients present
- Stress is low

Catabolism

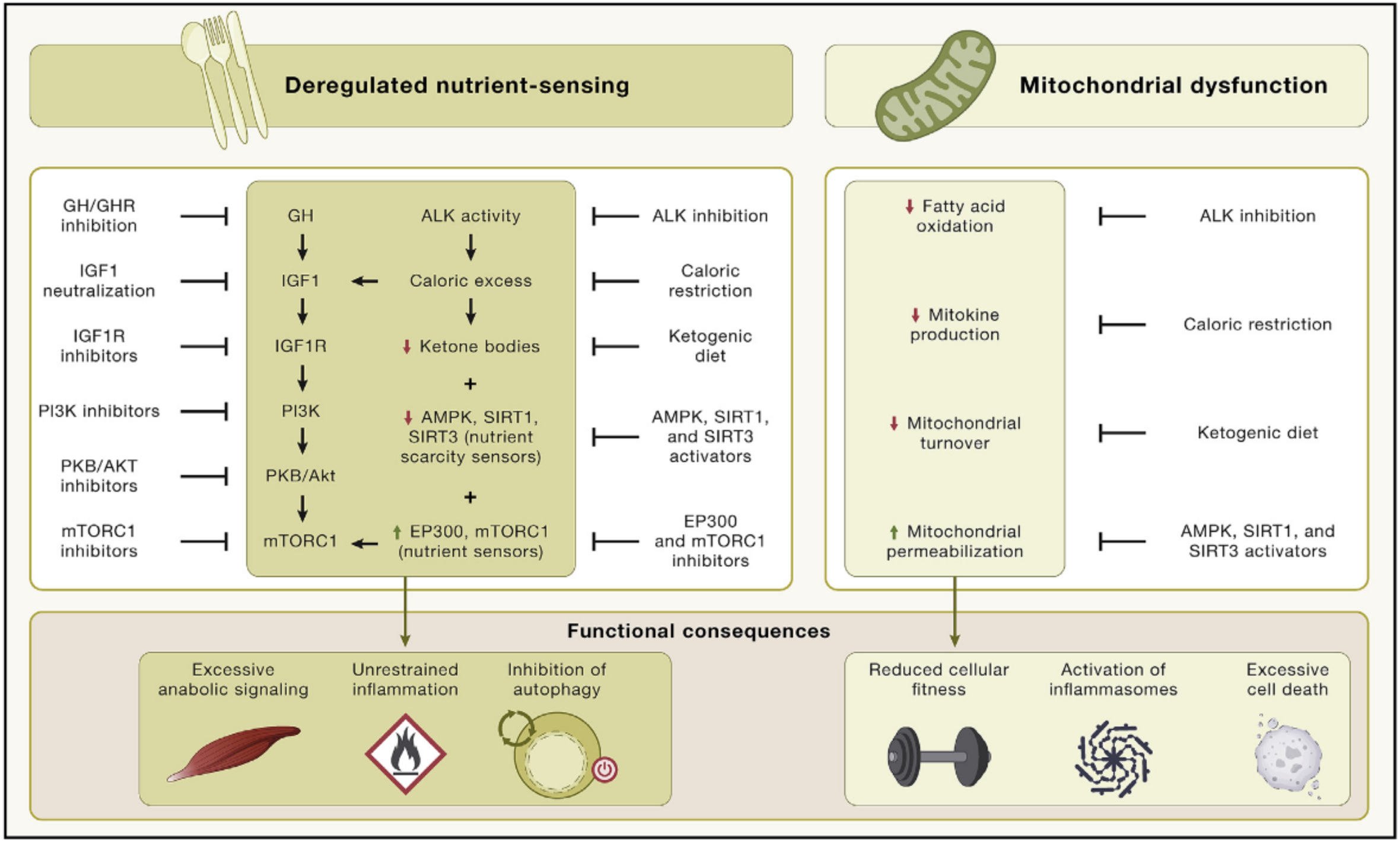
- Cellular defense mode
- Nutrient shortage
- Stress is present

Youth – network promotes beneficial anabolic responses

Adulthood – pro-aging properties

Deregulated Nutrient-Sensing

- Diet – practical intervention
- Overnutrition
- Caloric restriction
- Intermittent fasting
- Ketogenic diet
- Is this the best approach?


Deregulated nutrient-sensing
Mitochondrial dysfunction
GH/GHR inhibition
IGF1 neutralization
IGF1R inhibitors
PI3K inhibitors
PKB/AKT inhibitors
mTORC1 inhibitors
GH
IGF1
IGF1R
PI3K
PKB/Akt
mTORC1
ALK activity
Caloric excess
↓ Ketone bodies
+
↓ AMPK, SIRT1, SIRT3 (nutrient scarcity sensors)
+
↑ EP300, mTORC1 (nutrient sensors)
ALK inhibition
Caloric restriction
Ketogenic diet
AMPK, SIRT1, and SIRT3 activators
EP300 and mTORC1 inhibitors
↓ Fatty acid oxidation
↓ Mitokine production
↓ Mitochondrial turnover
↑ Mitochondrial permeabilization
ALK inhibition
Caloric restriction
Ketogenic diet
AMPK, SIRT1, and SIRT3 activators
Functional consequences
Excessive anabolic signaling
Unrestrained inflammation
Inhibition of autophagy
Reduced cellular fitness
Activation of inflammasomes
Excessive cell death

Mitochondrial Dysfunction

Two Functions

- Cellular powerhouse
- Also latent trigger of inflammation and cell death
- Reactive oxygen species or mitochondrial DNA leak out
- Activates the inflammasome

Mitochondrial function declines with age

Membranes become more permeable

Considered life threatening

Mitochondrial Dysfunction

The L-carnitine observation
Positive effects of L-carnitine supplementation on both pre-frail subjects and elderly men
Counteracting age-related declining L-carnitine levels
Optimizes fatty acid oxidation by the mitochondria
L-carnitine in optimal amounts is derived from adequate dietary protein intake

Cellular Senescence

Secondary to acute or chronic damage

Accumulates in multiple tissues at different rates

Mainly affects fibroblasts, endothelial cells and immune cells

- All cells can undergo

Linked to telomere shortening

Even found in the brain and heart

Cellular Senescence

Triggers

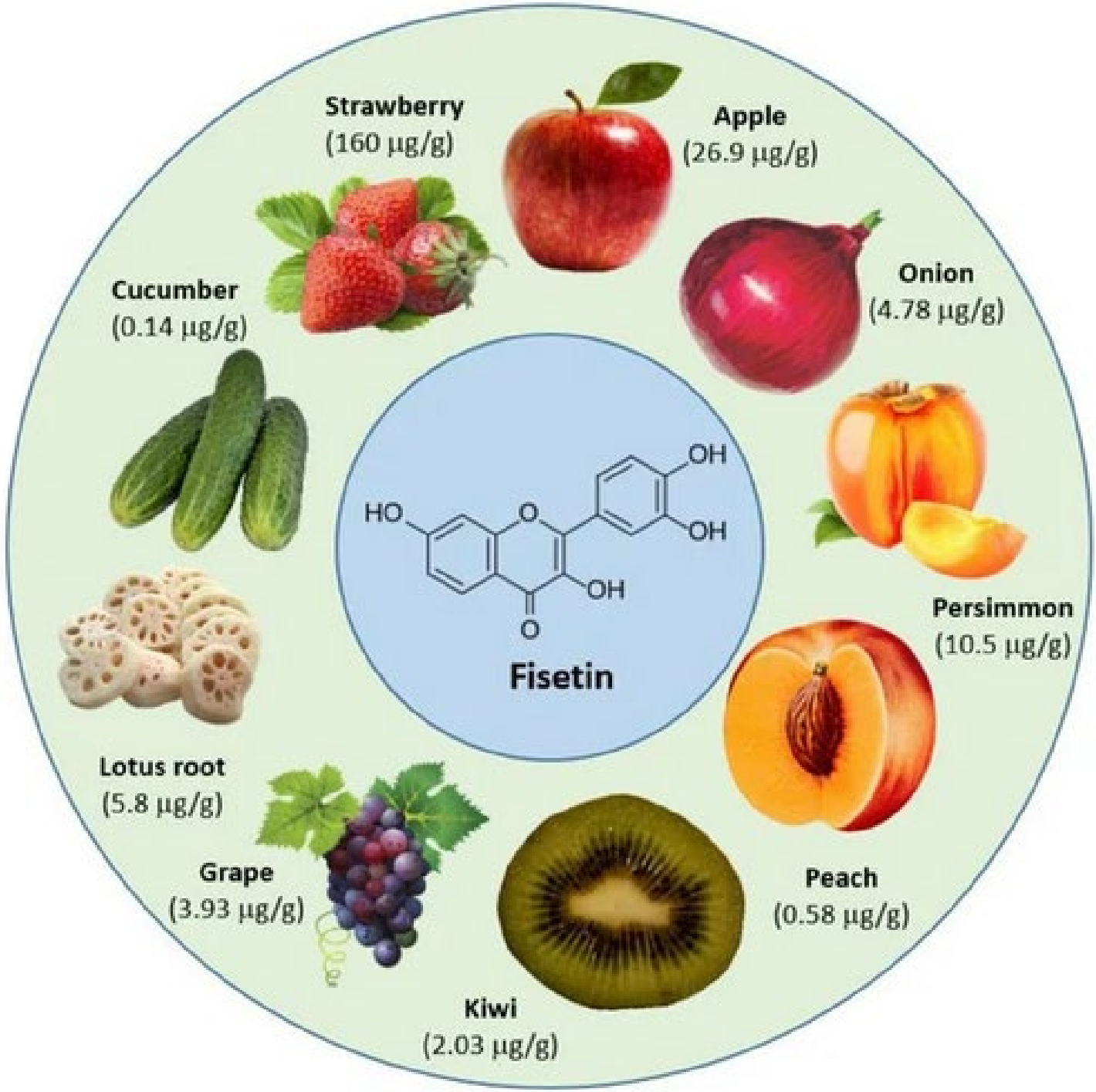
- Oncogenic signaling
- Genotoxic damage
- Critically short telomeres
- Mitochondrial damage
- Viral or bacterial infection
- Oxidative damage
- Nutrient imbalance
- Mechanical stress

Non-Proliferative Disease Associations

- Lung fibrosis
- Kidney diseases
- Liver steatosis
- Obesity-associated metabolic syndrome
- Type I and II diabetes
- Atherosclerosis
- Alzheimer’s & Parkinson’s diseases

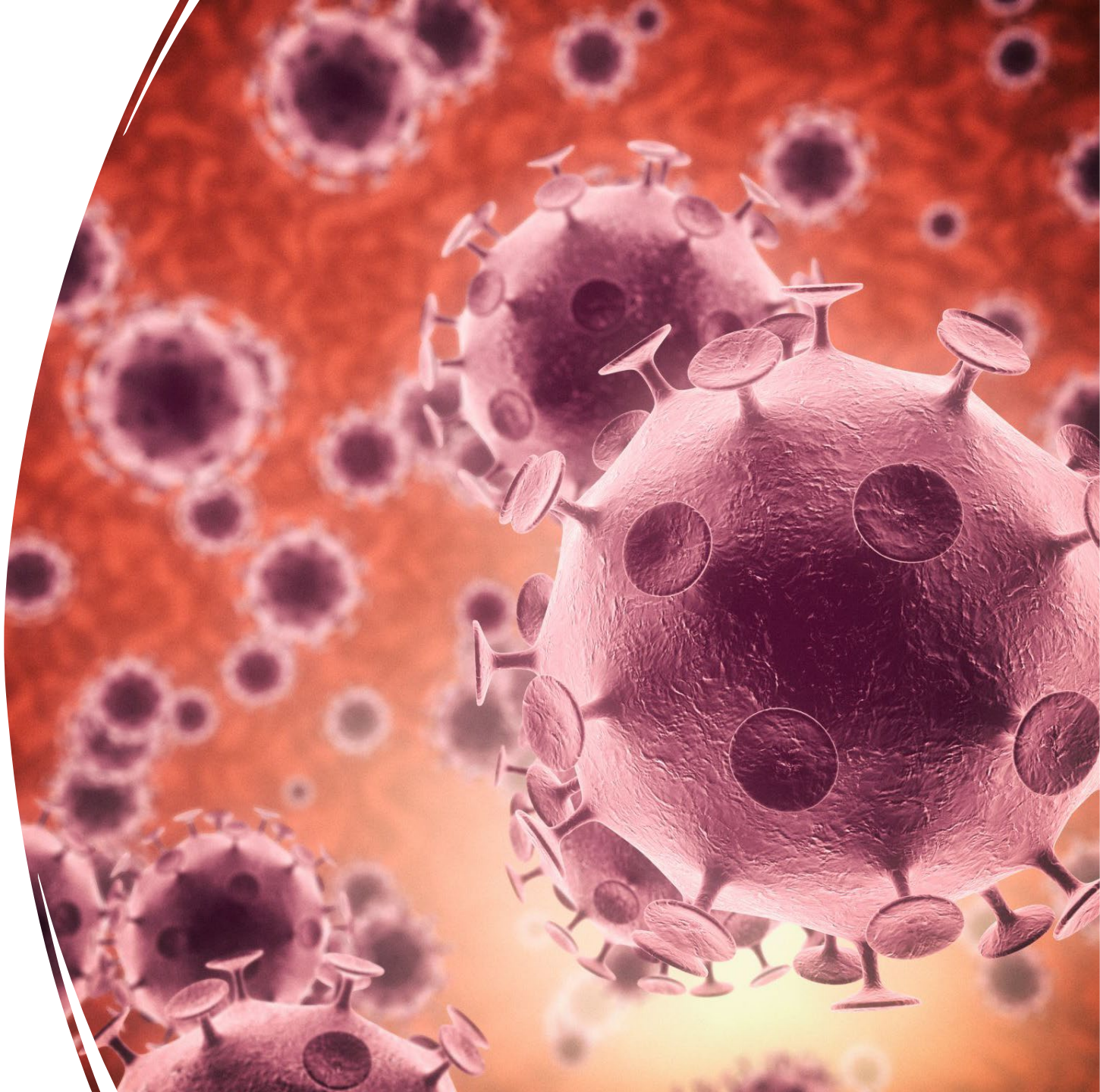
Cellular Senescence - Senolytics

- Senolytics
  - Chemical compounds that selectively kill senescent cells
  - Attempts to mimic the natural immune response
  - Natural flavonoid options
    - Quercetin
    - Fisetin
  - Drug options



Cellular Senescence

- Response to stress and damage
- Normally followed by immune clearance
- Upon aging or chronic damage, fails to be eliminated
- Becomes pathogenic due to inflammation and fibrosis



Immunosenescence

Thymus – Pillar of adaptive immunity

Starting in puberty, involution begins

Involution – loss of stromal cells and fat infiltration

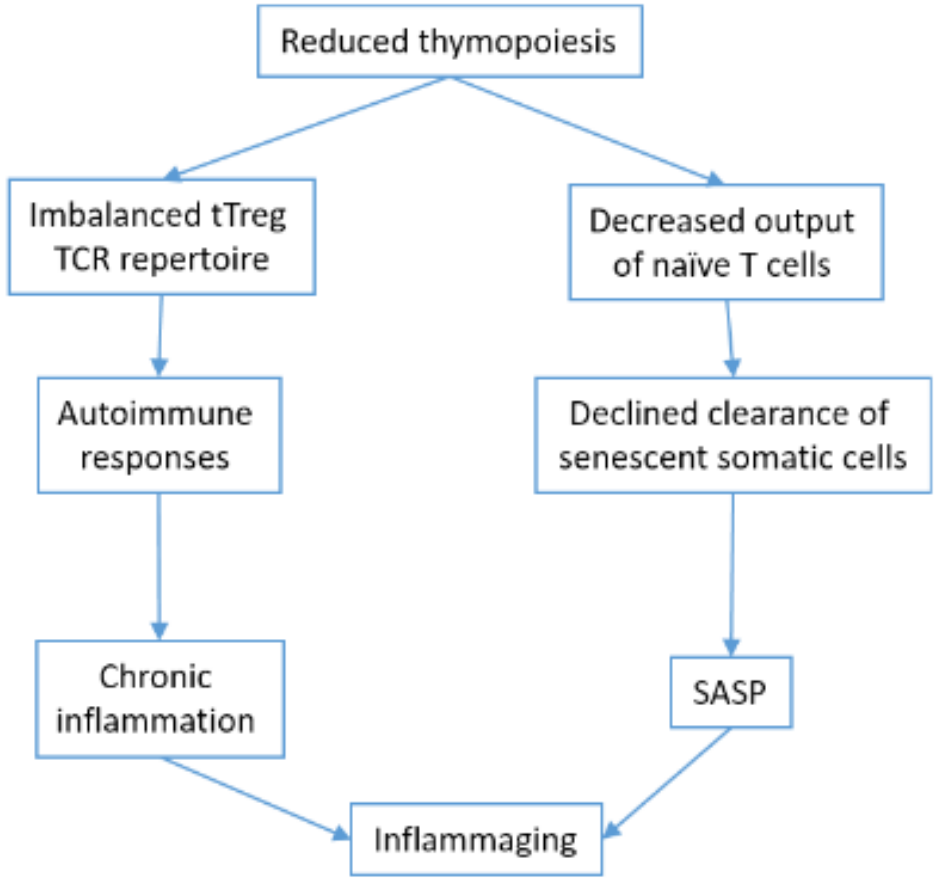
Decreases T cell response to new antigens

Cell-senescence associated T cells secrete pro-inflammatory cytokines

Increases pattern towards SASP (senescence associated secretory phenotype)

Could be part of the reason for dysfunction of immune clearance of senescent cells

Mechanism of inflammaging



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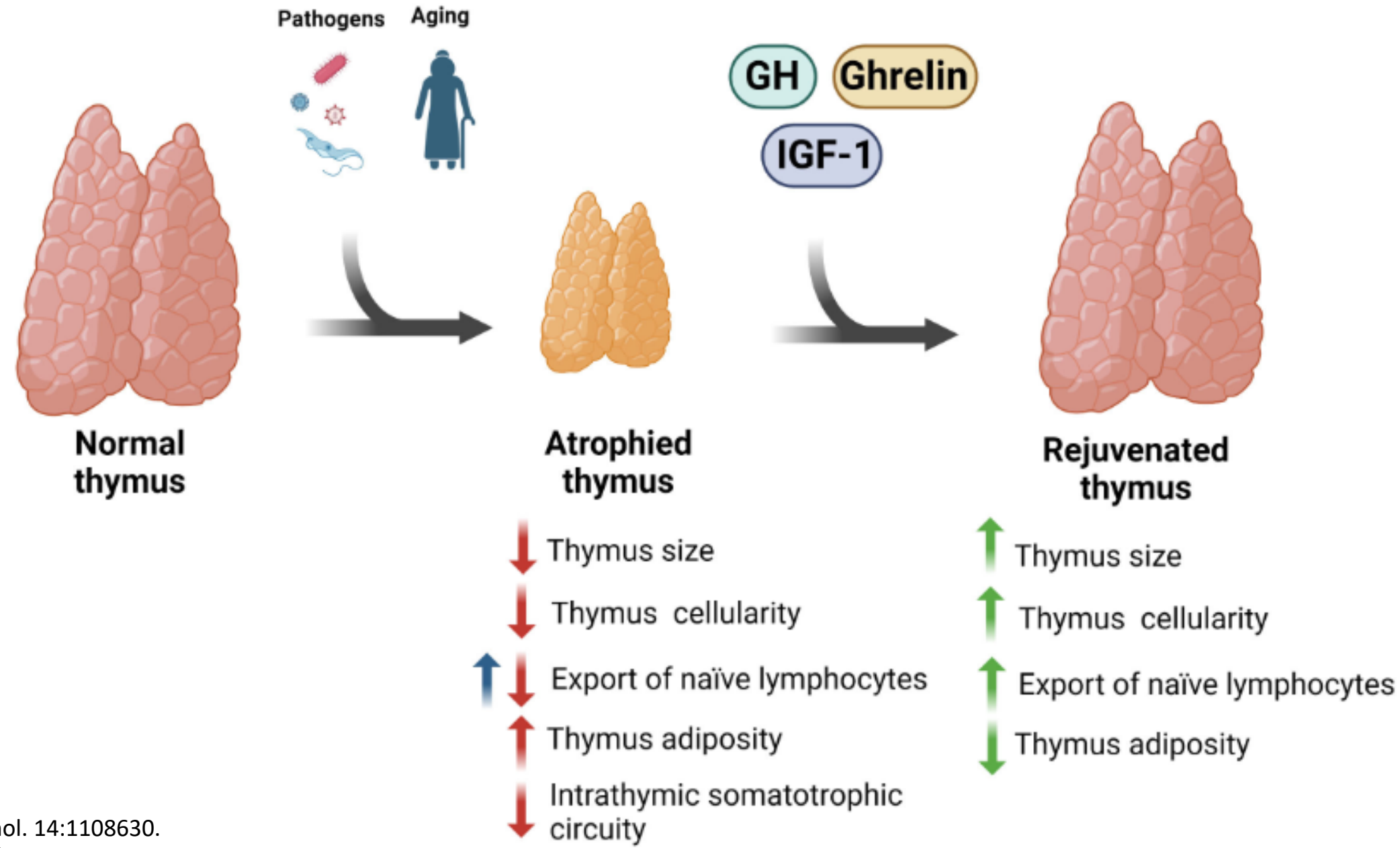

Front. Immunol. 14:1108630.
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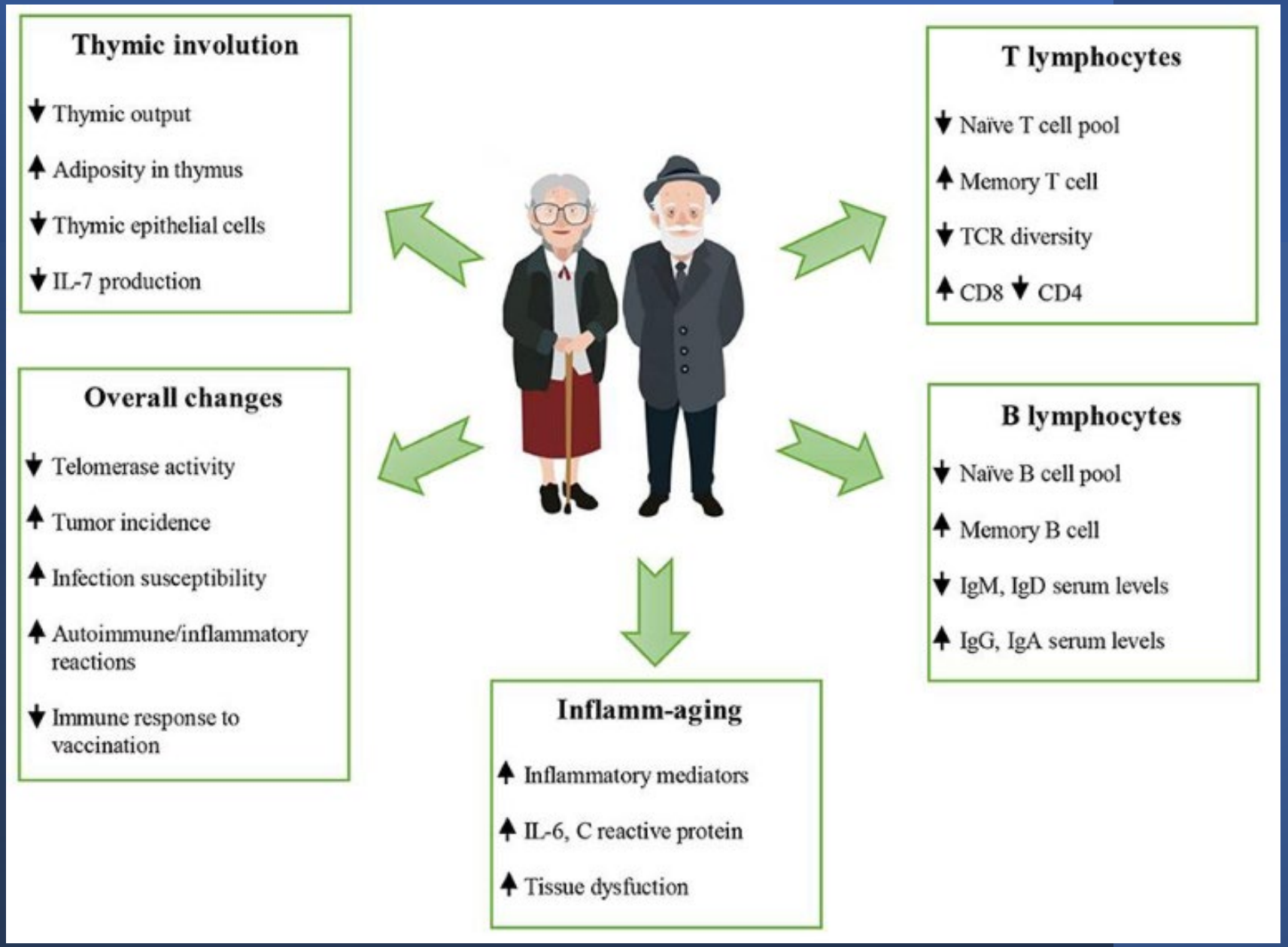
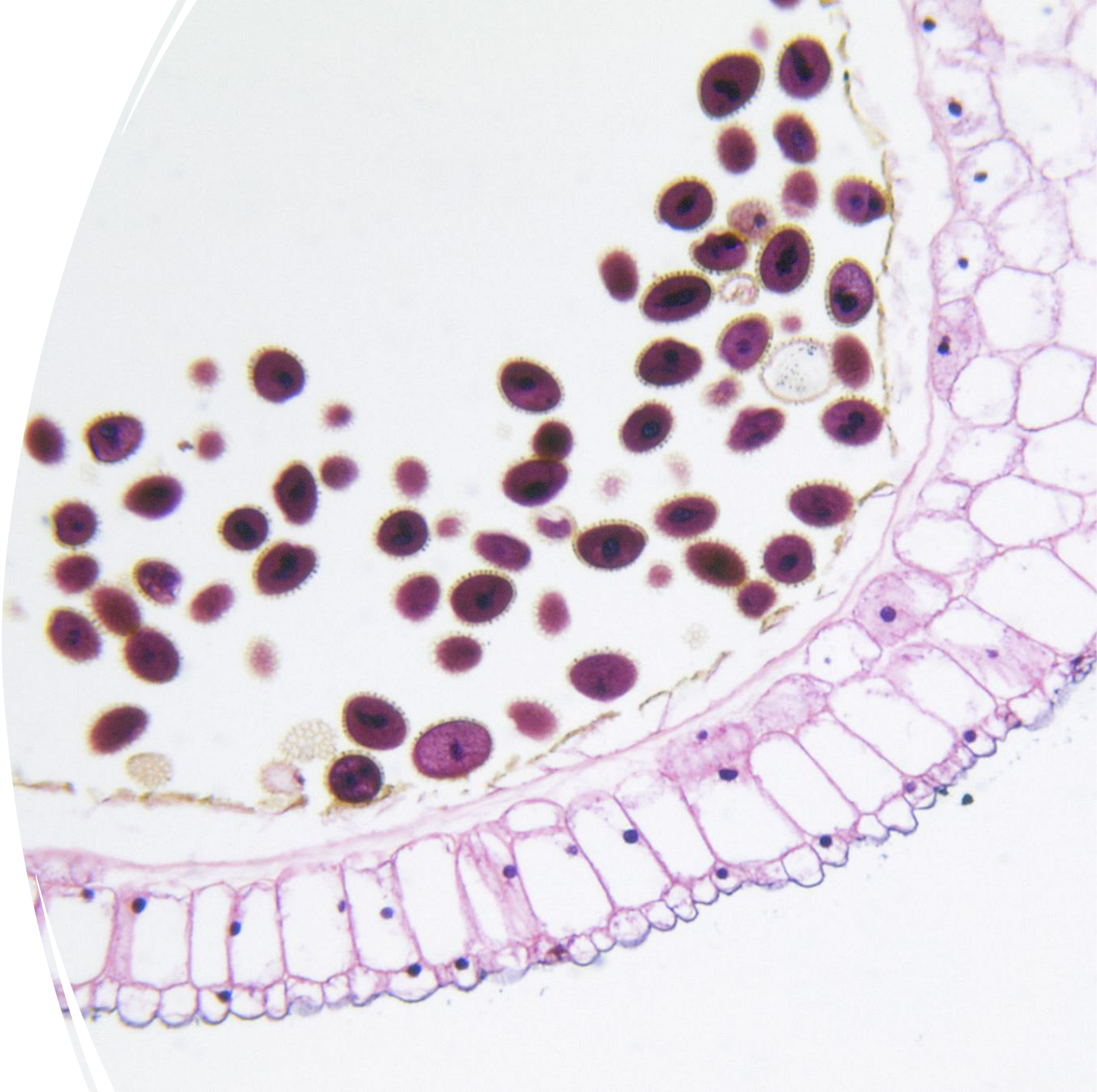

Thymic involution
Thymic output
Adiposity in thymus
Thymic epithelial cells
IL-7 production
T lymphocytes
Naïve T cell pool
Memory T cell
TCR diversity
CD8 CD4
Overall changes
Telomerase activity
Tumor incidence
Infection susceptibility
Autoimmune/inflammatory reactions
Immune response to vaccination
B lymphocytes
Naïve B cell pool
Memory B cell
IgM, IgD serum levels
IgG, IgA serum levels
Inflamm-aging
Inflammatory mediators
IL-6, C reactive protein
Tissue dysfuction

Stem Cell Exhaustion

- Aging – reduced tissue renewal at steady state
  - Impaired tissue repair upon injury
- Stem cells – undifferentiated cells
- Ability to differentiate into cell types required for healing



Altered Intercellular Communication

Aging – progressive alterations in intercellular communication

More noise in the network

Compromises homeostasis

Deficiencies in neural, neuroendocrine and hormonal pathways

- Adrenergic
- Noradrenergic
- Insulin / IGF-1
- Renin-angiotensin
- Sex hormones

Altered Intercellular Communication

Associated with altered gut-brain interactions

Promoter of dysbiosis

Vitality in the blood

- Proaging bloodborne factors
- Antiaging bloodborne factors
- Molecules in the blood linked to age
- Crossover studies in mice to date
  - Young mice given old mice blood – induces features of aging
  - Old mice given young mice blood – restores repair capacity

Chronic Inflammation

Inflammaging – inflammation increases during aging

Systemic manifestations

Increased inflammatory cytokines

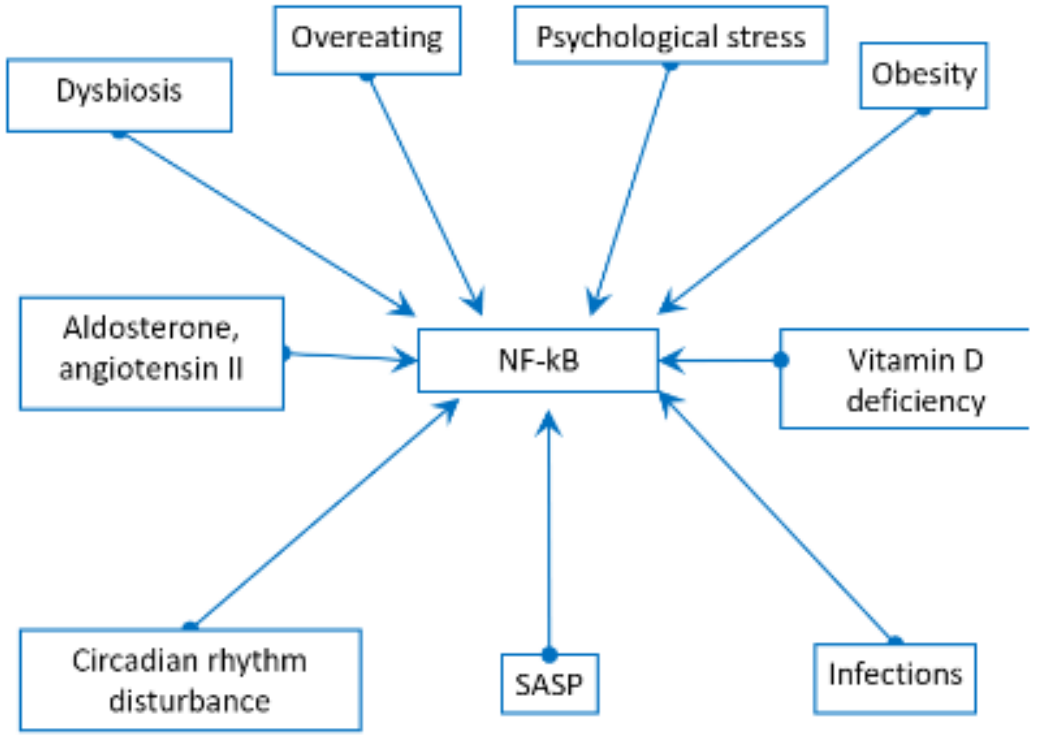
- IL-6 – predictor of all cause mortality
- Correlates with CRP

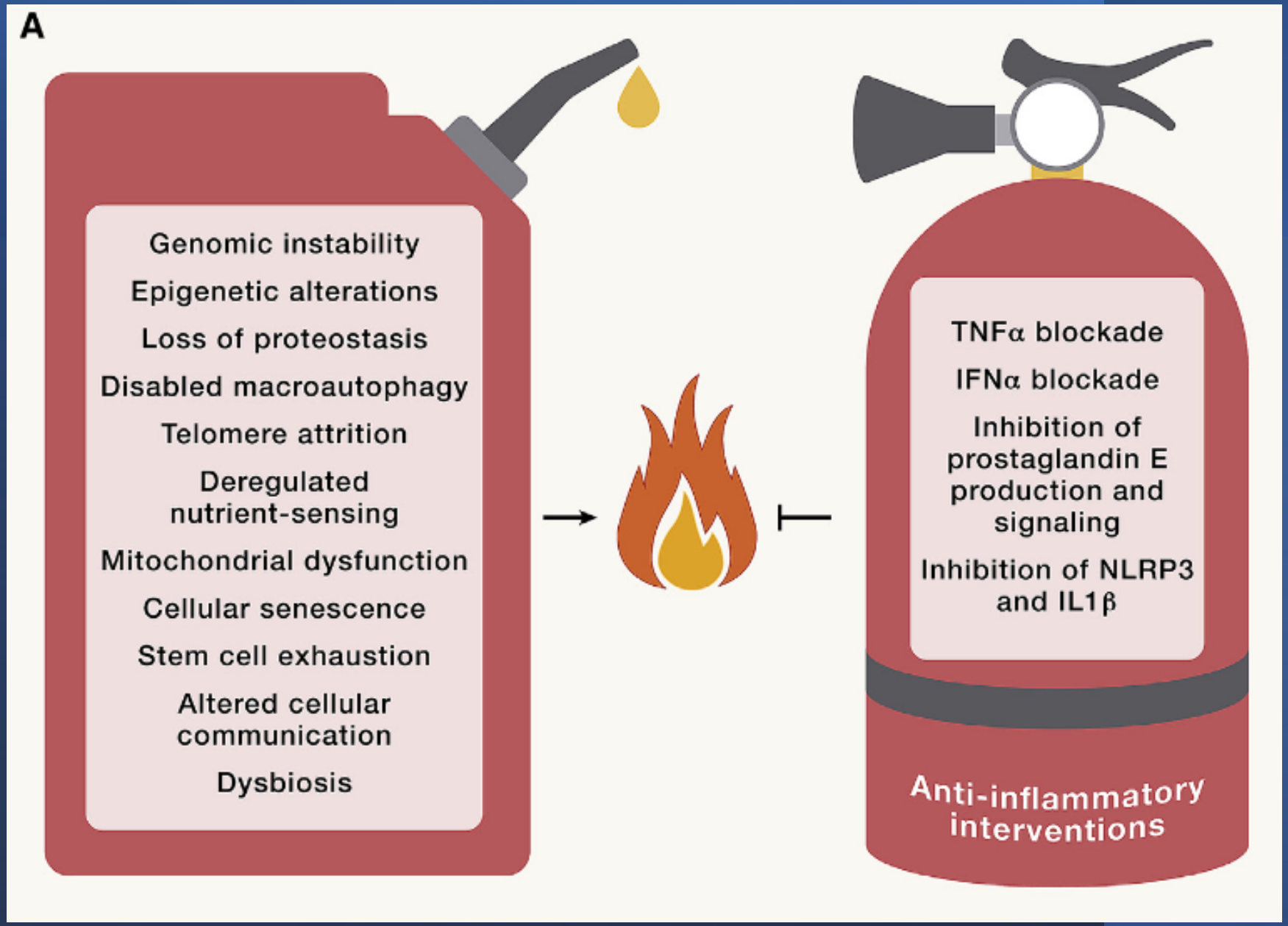
Increased inflammation decreases immune function

- Increases in TH1 and TH17 cells
  - Defective immunosurveillance
  - Loss of self-tolerance
  - Reduced maintenance of biological barriers (gut, brain)

Chronic Inflammation

- Byproduct of all other hallmarks of aging
- Exacerbated by disturbances of circadian rhythms
- Target directly and indirectly
  - Directly – target TNF-α,NF-Kβ, inflammasome
  - Indirectly – address aforementioned areas of dysfunction

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Deregulated nutrient-sensing
Mitochondrial dysfunction
Cellular senescence
Stem cell exhaustion
Altered cellular communication
Dysbiosis
TNFα blockade
IFNα blockade
Inhibition of prostaglandin E production and signaling
Inhibition of NLRP3 and IL1β
Anti-inflammatory interventions

Dysbiosis

Disruption of the bacteria-host bidirectional communication

Associated conditions:

- Obesity
- Type II diabetes
- Ulcerative colitis
- Neurological disorders
- Cardiovascular disease
- Cancer

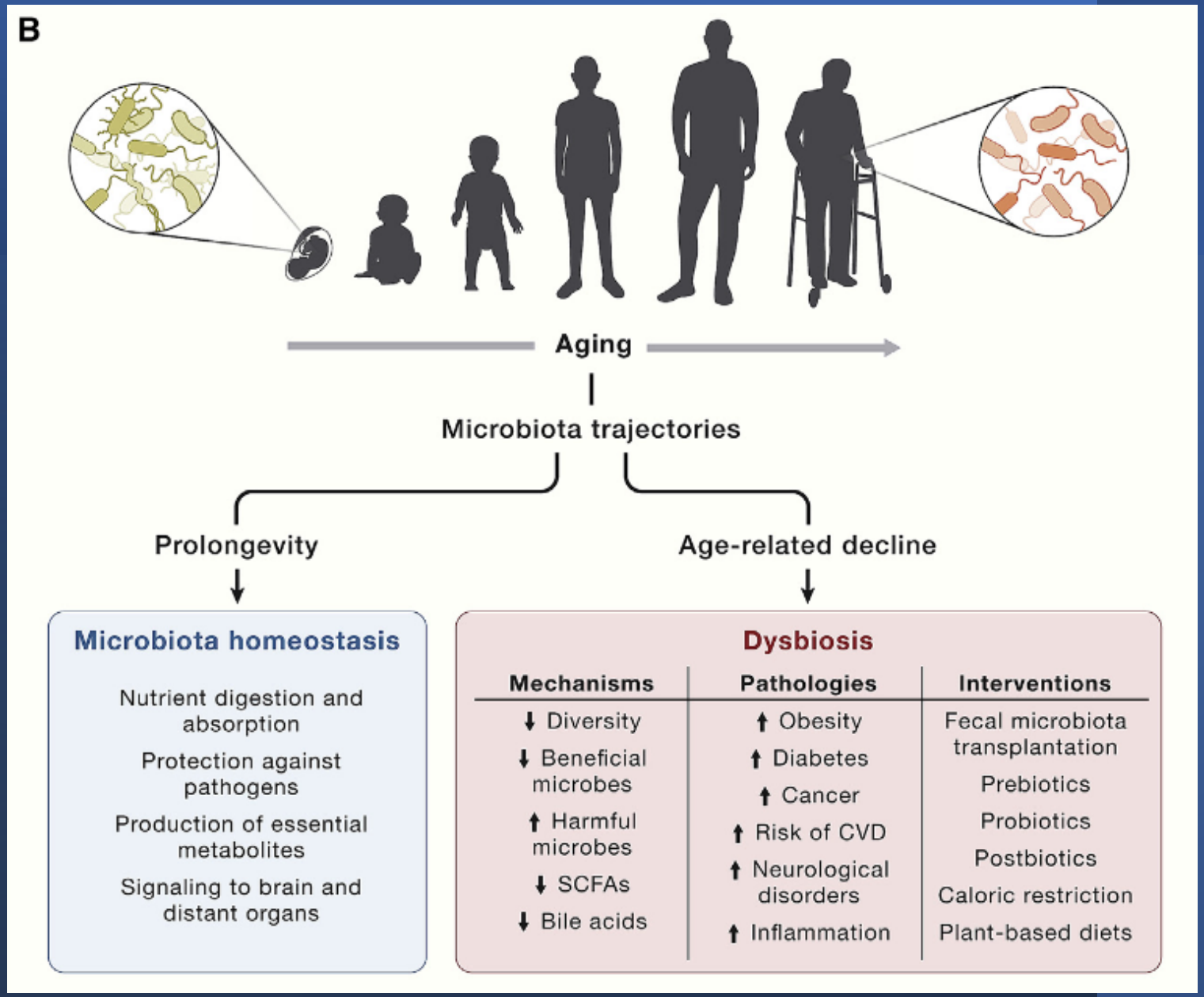
Dysbiosis

Intestinal community within the intestinal tract is highly variable

- Ethnicity
- Dietary factors
- Lifestyle habits
- Environmental conditions

Microbial profiles changes with aging

- Initial establishment in childhood
- Relatively stable throughout adulthood
- Gradual changes with aging
- Higher levels of *Akkermansia* and *Lactobacillus plantarum* are more advantageous


B
Aging
Microbiota trajectories
Prolongevity
Age-related decline
Microbiota homeostasis
Nutrient digestion and absorption
Protection against pathogens
Production of essential metabolites
Signaling to brain and distant organs
Dysbiosis
Mechanisms
↓ Diversity
↓ Beneficial microbes
↑ Harmful microbes
↓ SCFAs
↓ Bile acids
Pathologies
↑ Obesity
↑ Diabetes
↑ Cancer
↑ Risk of CVD
↑ Neurological disorders
↑ Inflammation
Interventions
Fecal microbiota transplantation
Prebiotics
Probiotics
Postbiotics
Caloric restriction
Plant-based diets

12 Hallmarks of Aging

- This is your future
- Appears ominous
- Do you have to accept it?

8 Hallmarks of Health

Spatial Compartmentalization

- Integrity of barriers
- Containment of perturbations

Maintenance of homeostasis

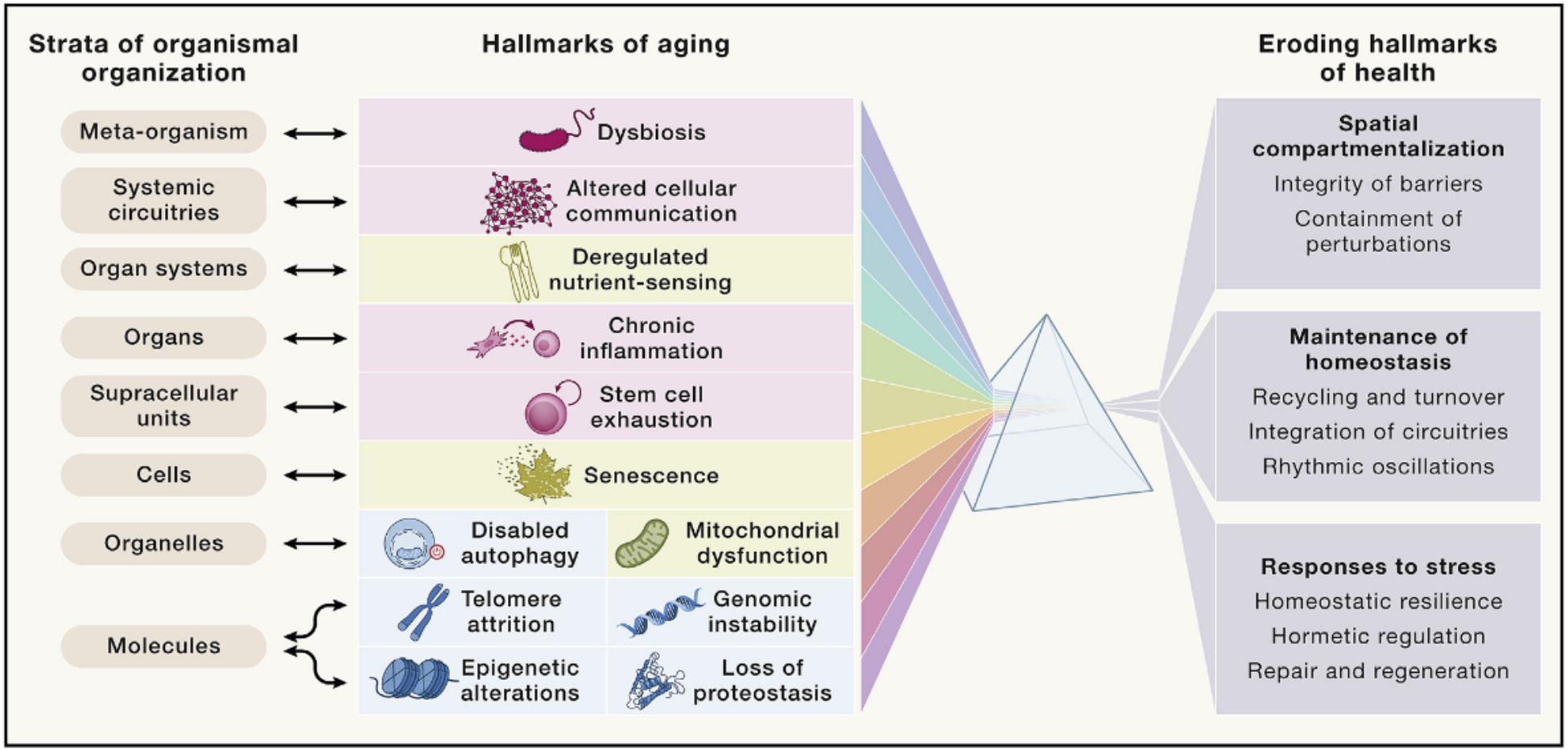
- Recycling and turnover
- Integration of circuitries
- Rhythmic oscillations

Response to Stress

- Homeostatic resilience
- Hormetic regulation
- Repair and regeneration

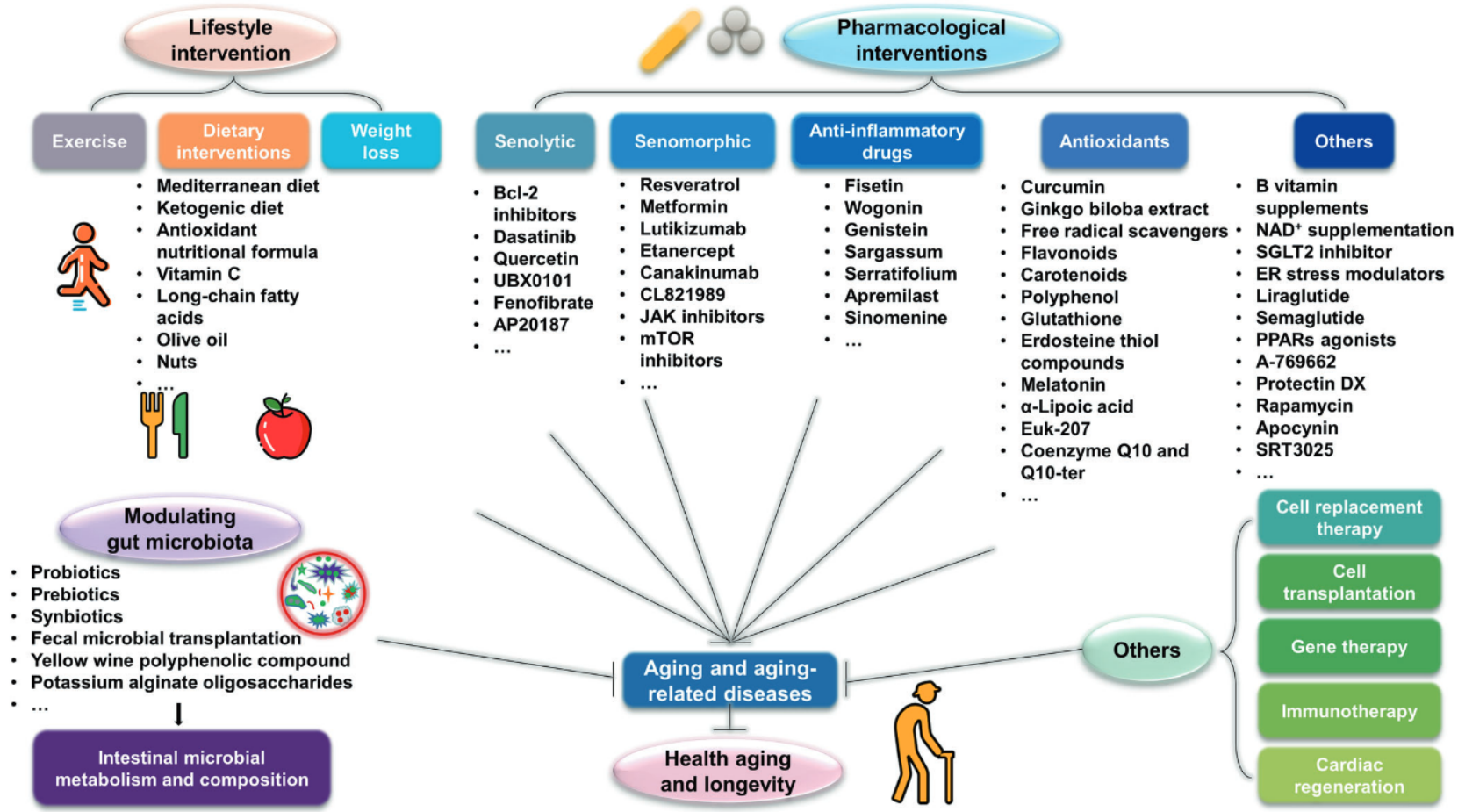
Hallmarks of Health

Cell 186, January 19, 2023



Intervention Against Aging

Signal Transduction and Targeted Therapy (2022) 7:391



Caloric Restriction

Premise – Reduced expression of DNA repair enzymes

Caloric restriction – maintains integrity of the genome

Primarily AMPK and Sirt1 stimulation

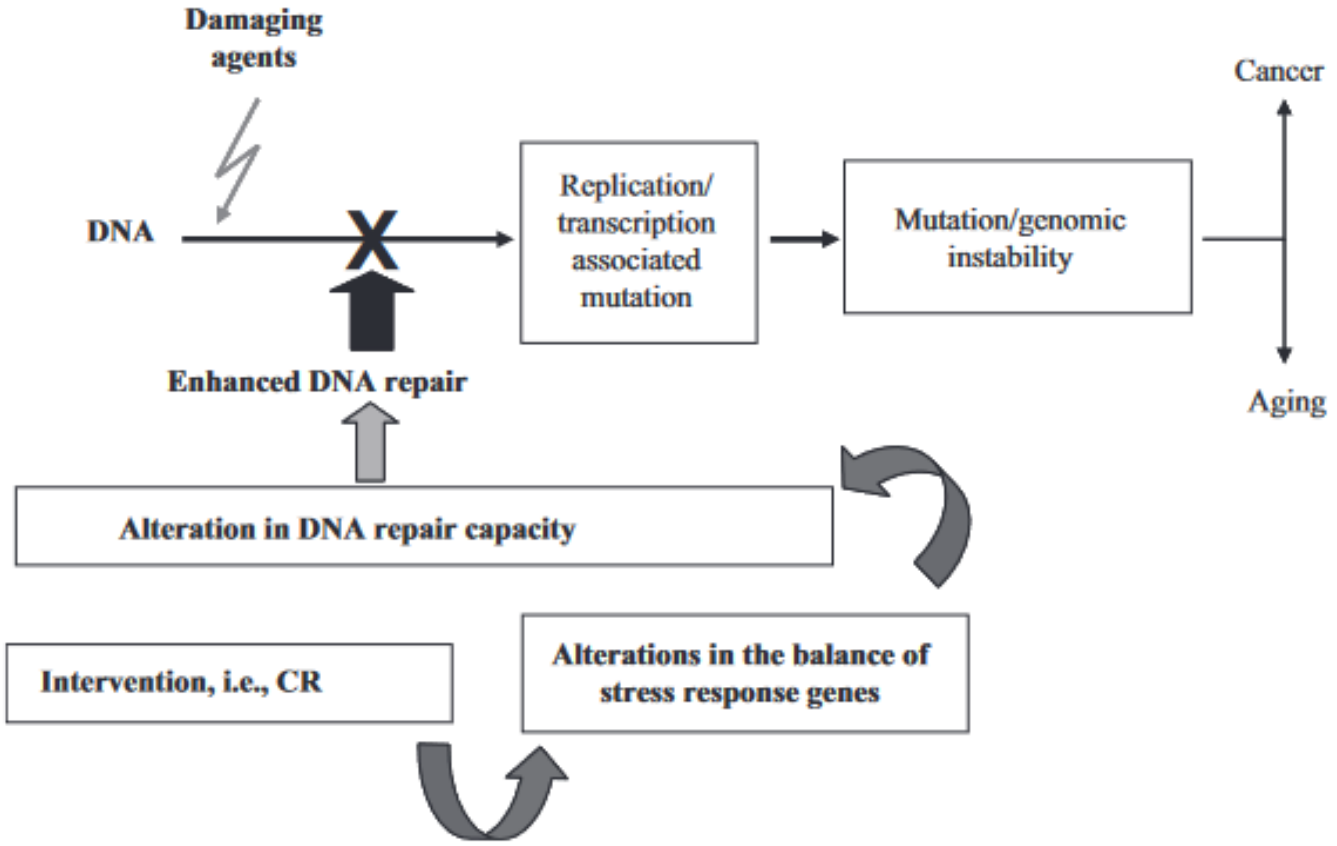
- Increased intracellular AMP and ADP

Also possible with nutrients and exercise

Stimulation of autophagy

Energetic Stress = autophagy

Elevated calcium, NAD+ and ROS instigate autophagy

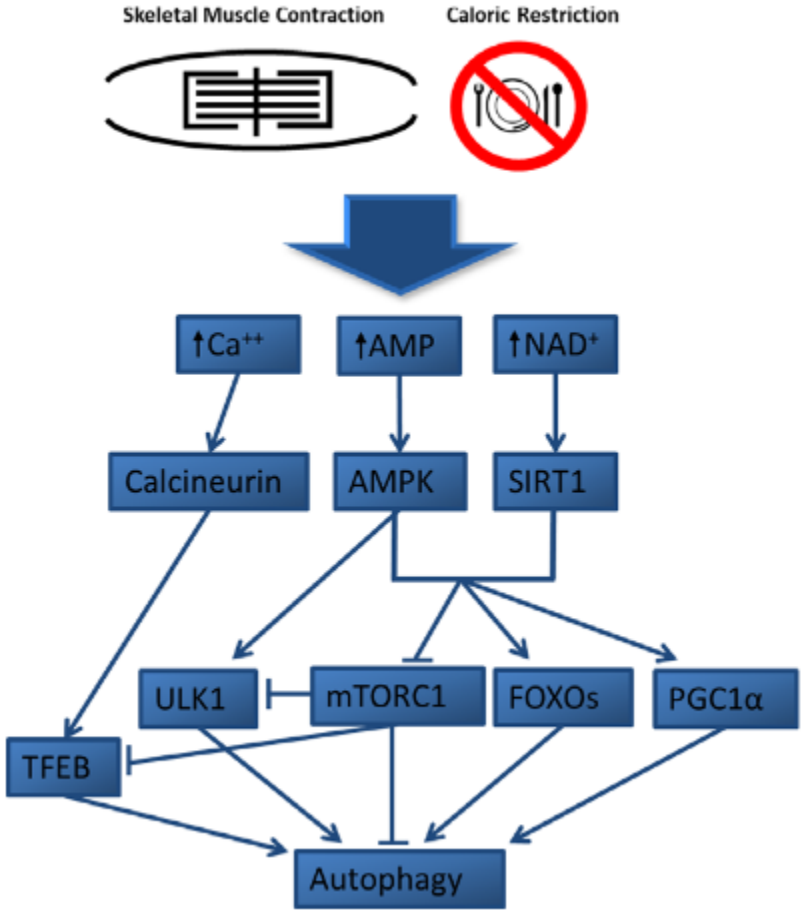


Nucleic Acids Research, 2007, Vol. 35, No. 22 7485–7496

Exercise and Caloric Restriction

Caloric restriction

- Not to be applied universally
- Ideal with metabolic derangement
- Not ideal with low cortisol levels
- Cortisol – DHEA-S ratio
- Random vs. intermittent vs. time-restricted eating



Micronutrient Stability

Diet as a determinant of genomic stability

Influences

- Dietary carcinogens
- Activation / Detoxification of carcinogens
- DNA Repair
- DNA Synthesis
- Apoptosis

Current dietary RDAs are insufficient

- Based on overt disease prevention – vitamin C and scurvy
- Not based on preventing nuclear and mitochondrial DNA

Nutrients for Genomic Stability

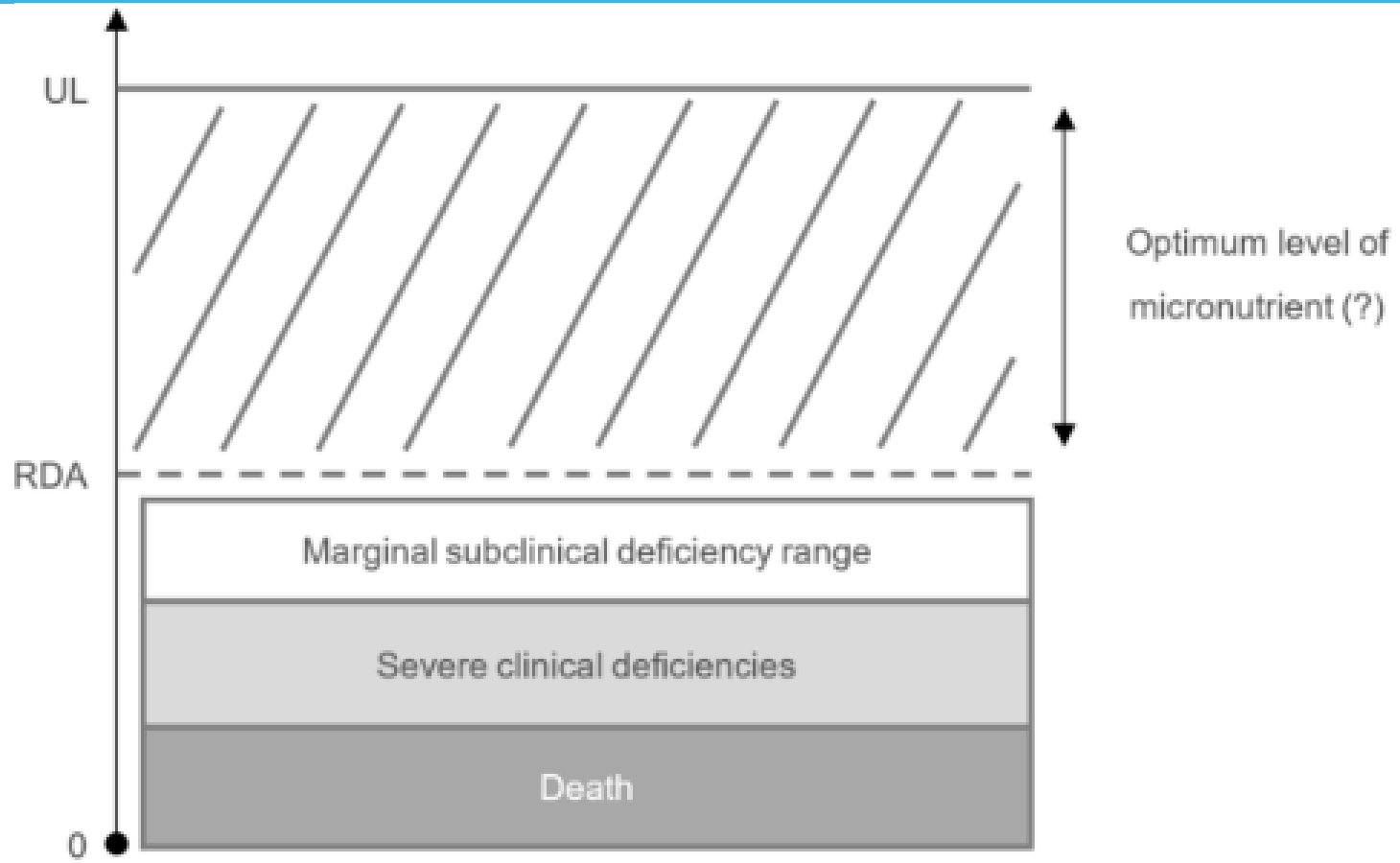
- Two types of deficiencies, marginal (borderline) and overt (functional)
- Marginal deficiencies impact damage and repair rate
- Folate
- B12
- Niacin
- Zinc
- Macrocytic anemia is an indicator of deficiencies significant enough to suggest genomic instability (DNA methylation)

Mutat Res. 2001 Apr 18;475(1-2):1-6.

Food Chem Toxicol. 2002 Aug;40(8):1113-7.

RDA Limitations

- For optimal immune protection and resistance to infection, daily intakes may need to be much higher than the RDAs
- Roughly 25–75% of people have a dietary intake that is less than the RDA, depending on the micronutrient

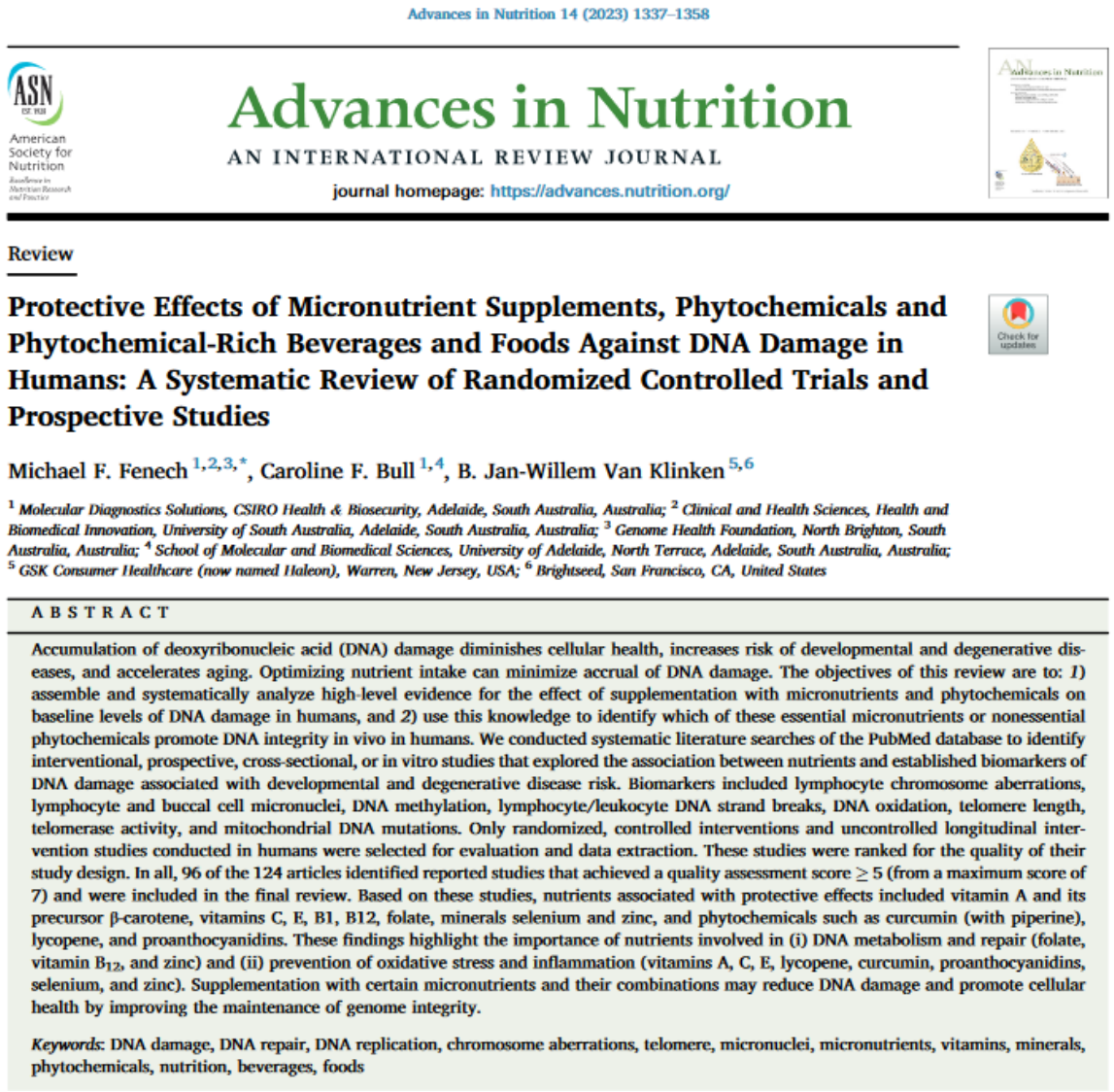


Nutrients 2020, 12, 236

Nutrient Deficiencies

- Major cause of DNA damage in humans
- Vitamins and minerals
  - Substrates
  - Cofactors in DNA replication and repair
  - Aid neutralization of genotoxins (exogenous and endogenous)
- Vitamins C, E, K
- Various polyphenols
- Folate, B2, B3, B6, B12
- Zinc, iron, magnesium, manganese, calcium, selenium

Advances in Nutrition 14 (2023) 1337–1358



American Society for Nutrition

Advances in Nutrition
AN INTERNATIONAL REVIEW JOURNAL
journal homepage: https://advances.nutrition.org/

Review

Protective Effects of Micronutrient Supplements, Phytochemicals and Phytochemical-Rich Beverages and Foods Against DNA Damage in Humans: A Systematic Review of Randomized Controlled Trials and Prospective Studies

Michael F. Fenech [1,2,3,*], Caroline F. Bull [1,4], B. Jan-Willem Van Klinken [5,6]

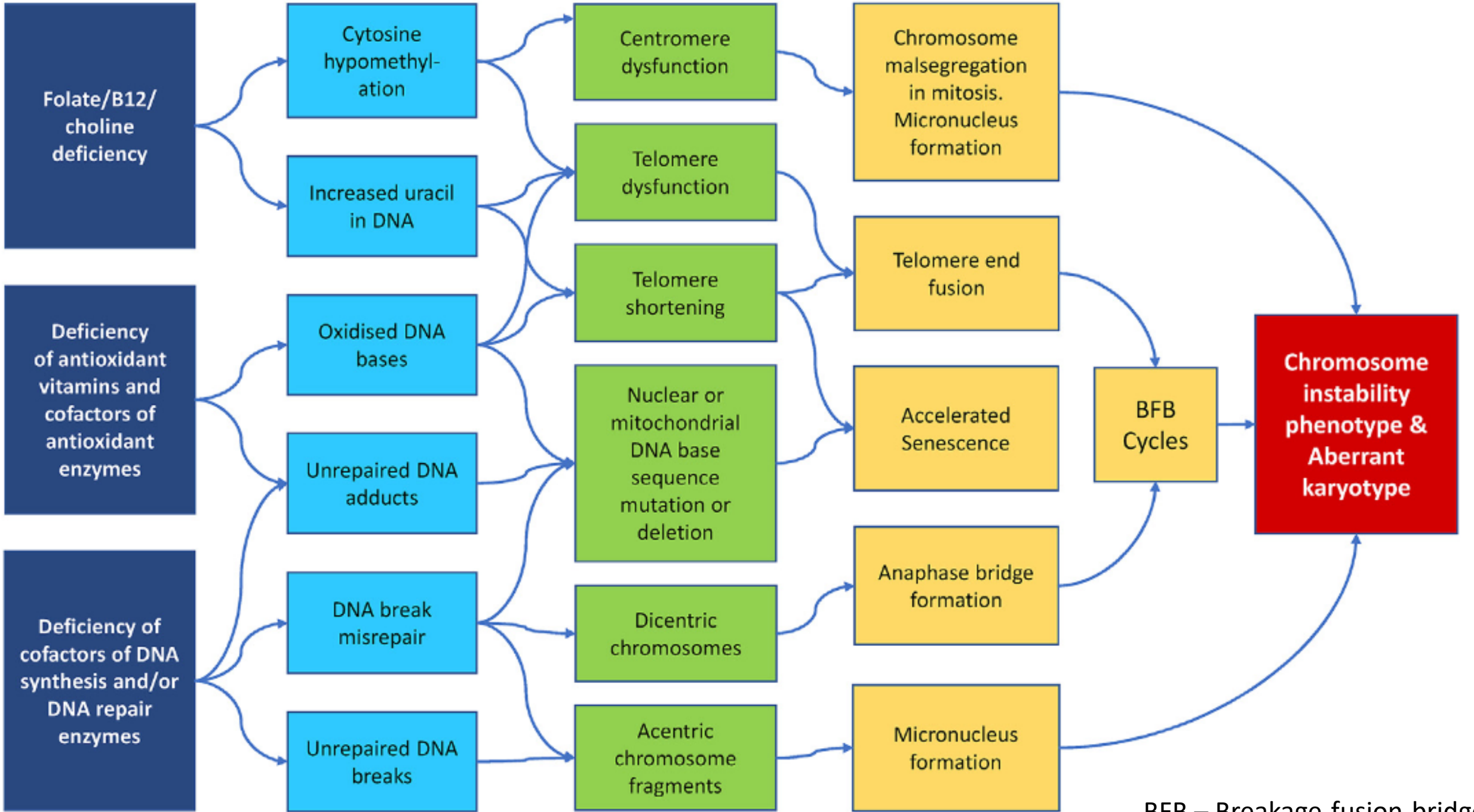
[1] *Molecular Diagnostics Solutions, CSIRO Health & Biosecurity, Adelaide, South Australia, Australia;* [2] *Clinical and Health Sciences, Health and Biomedical Innovation, University of South Australia, Adelaide, South Australia, Australia;* [3] *Genome Health Foundation, North Brighton, South Australia, Australia;* [4] *School of Molecular and Biomedical Sciences, University of Adelaide, North Terrace, Adelaide, South Australia, Australia;* [5] *GSK Consumer Healthcare (now named Haleon), Warren, New Jersey, USA;* [6] *Brightseed, San Francisco, CA, United States*

ABSTRACT

Accumulation of deoxyribonucleic acid (DNA) damage diminishes cellular health, increases risk of developmental and degenerative diseases, and accelerates aging. Optimizing nutrient intake can minimize accrual of DNA damage. The objectives of this review are to: *1)* assemble and systematically analyze high-level evidence for the effect of supplementation with micronutrients and phytochemicals on baseline levels of DNA damage in humans, and *2)* use this knowledge to identify which of these essential micronutrients or nonessential phytochemicals promote DNA integrity in vivo in humans. We conducted systematic literature searches of the PubMed database to identify interventional, prospective, cross-sectional, or in vitro studies that explored the association between nutrients and established biomarkers of DNA damage associated with developmental and degenerative disease risk. Biomarkers included lymphocyte chromosome aberrations, lymphocyte and buccal cell micronuclei, DNA methylation, lymphocyte/leukocyte DNA strand breaks, DNA oxidation, telomere length, telomerase activity, and mitochondrial DNA mutations. Only randomized, controlled interventions and uncontrolled longitudinal intervention studies conducted in humans were selected for evaluation and data extraction. These studies were ranked for the quality of their study design. In all, 96 of the 124 articles identified reported studies that achieved a quality assessment score ≥ 5 (from a maximum score of 7) and were included in the final review. Based on these studies, nutrients associated with protective effects included vitamin A and its precursor β-carotene, vitamins C, E, B1, B12, folate, minerals selenium and zinc, and phytochemicals such as curcumin (with piperine), lycopene, and proanthocyanidins. These findings highlight the importance of nutrients involved in (i) DNA metabolism and repair (folate, vitamin B_{12}, and zinc) and (ii) prevention of oxidative stress and inflammation (vitamins A, C, E, lycopene, curcumin, proanthocyanidins, selenium, and zinc). Supplementation with certain micronutrients and their combinations may reduce DNA damage and promote cellular health by improving the maintenance of genome integrity.

Keywords: DNA damage, DNA repair, DNA replication, chromosome aberrations, telomere, micronuclei, micronutrients, vitamins, minerals, phytochemicals, nutrition, beverages, foods

Advances in Nutrition 14 (2023) 1337-1358



BFB – Breakage-fusion-bridge

Guanine

- Low redox potential
- Most susceptible to oxidation
- Results in guanine lesions
  - 8-oxo-7,8-dihydro-2’-deoxyguanosine (8-oxodG)
  - 8-hydroxy-2’-deoxyguanosine (8-OHdG)
  - Formed 105 times per day per cell
- Malnutrition
  - ROS accumulation
  - Impaired repair efficiency
  - Subsequent mutations
  - Pathological conditions

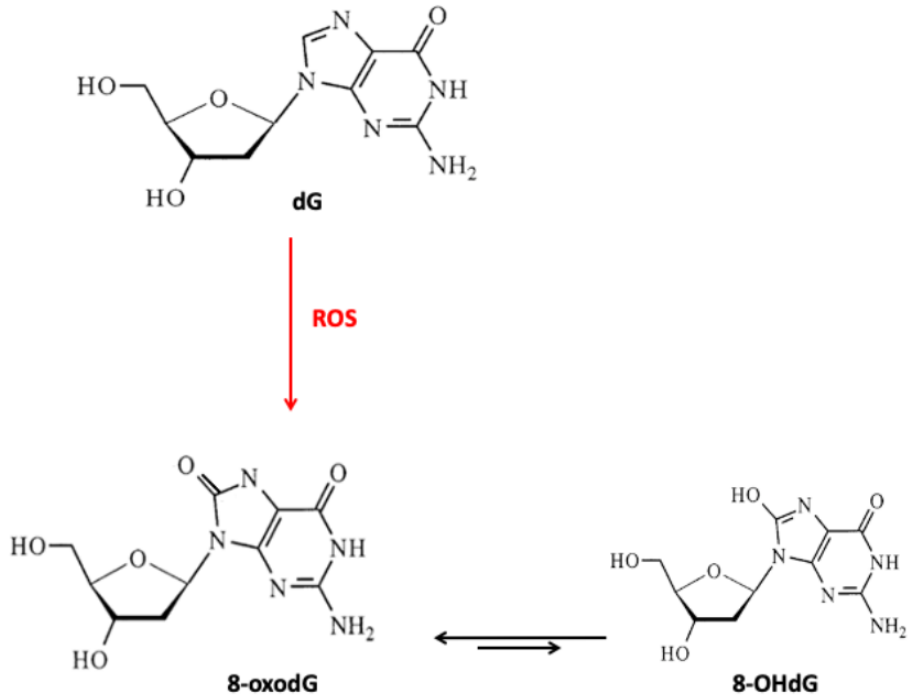
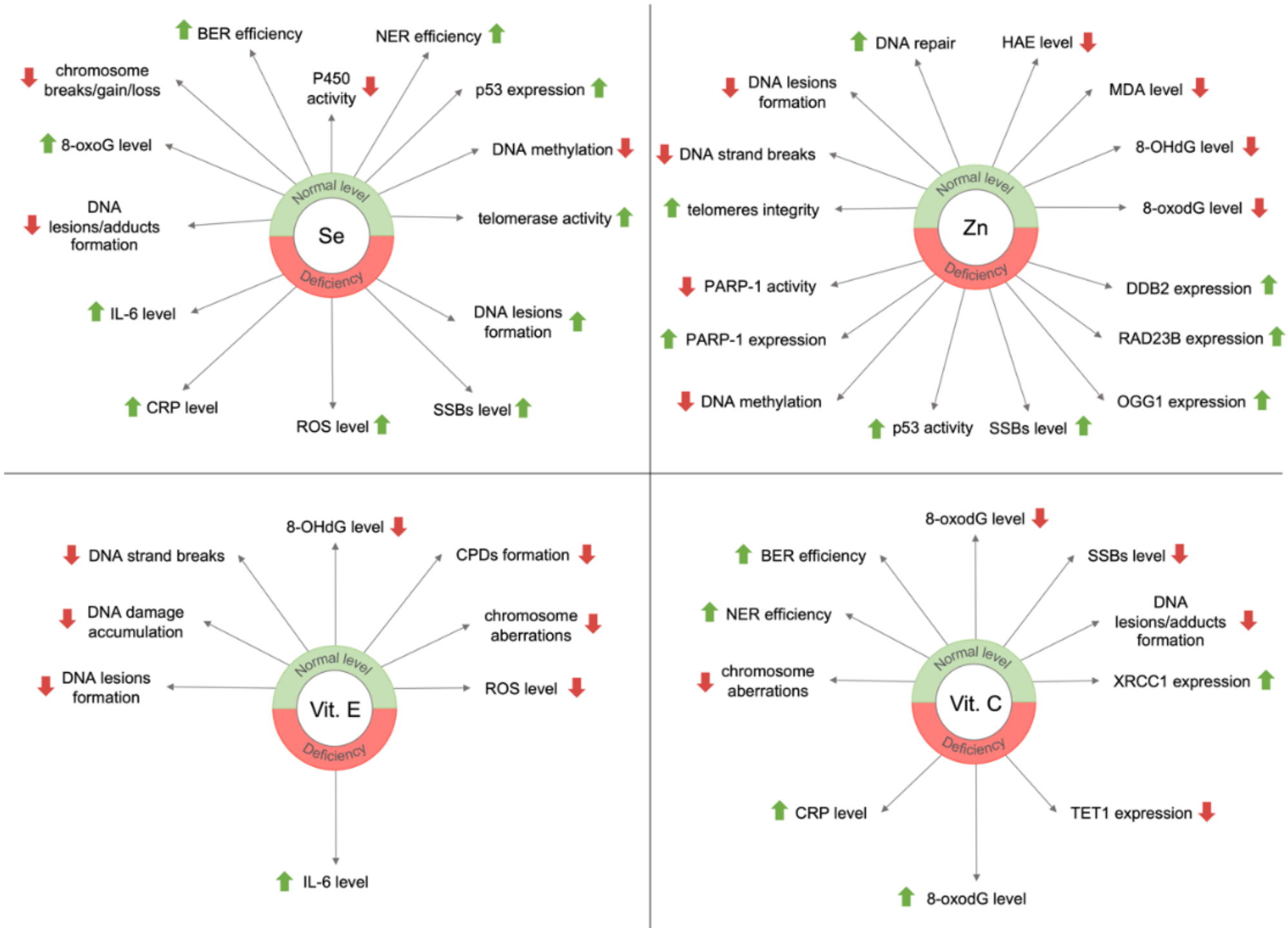


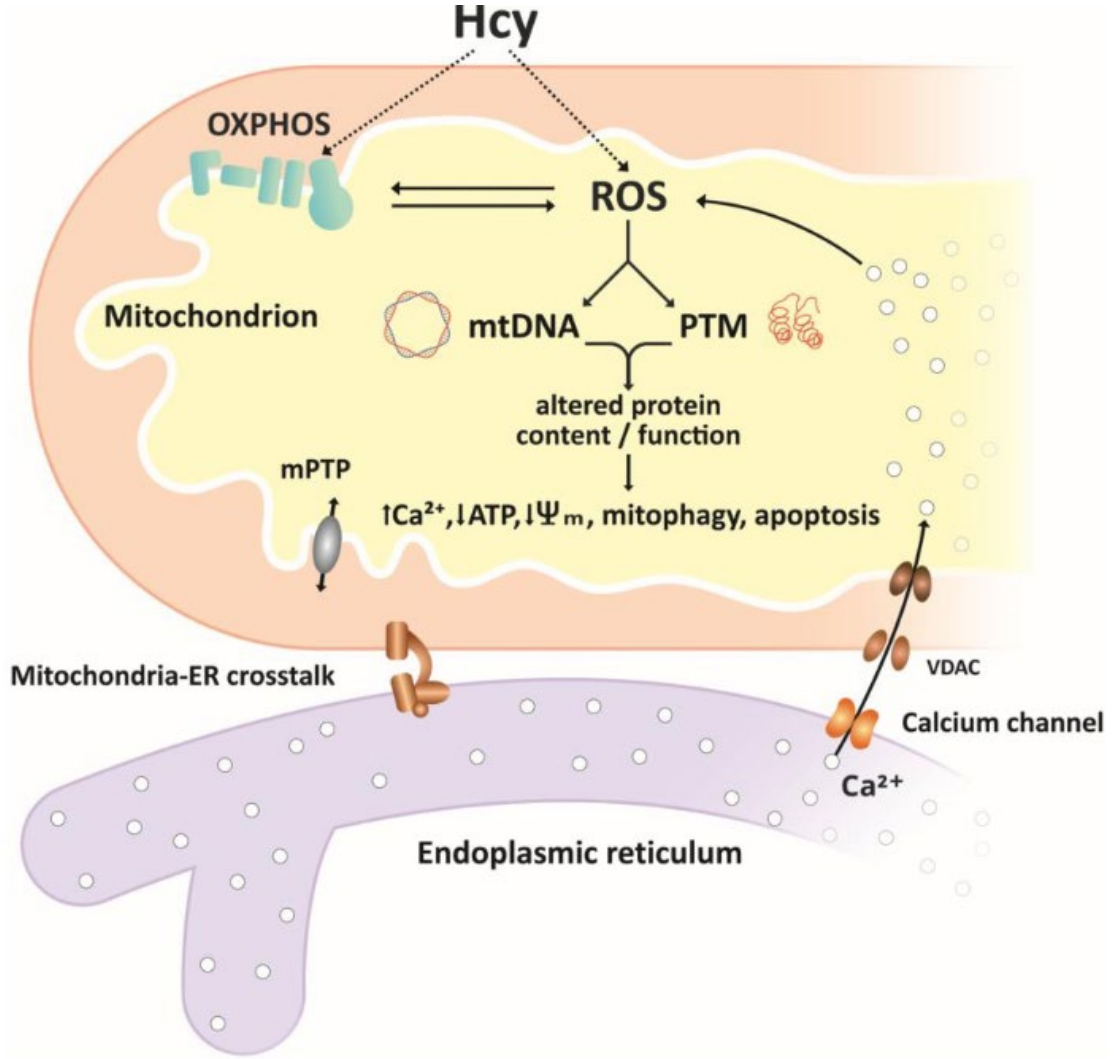
Figure 2. Guanosine and its oxidative modifications. dG, 2’-deoxyguanosine; 8-oxodG, 8-oxo-7, 8-dihydro-2’-deoxyguanosine; 8-OHdG, 8-hydroxy-2’-deoxyguanosine; ROS, reactive oxygen species.

Nutrients 2020, 12, 3364


BER efficiency
NER efficiency
chromosome breaks/gain/loss
P450 activity
p53 expression
8-oxoG level
DNA methylation
Normal level
Se
Deficiency
DNA lesions/adducts formation
telomerase activity
IL-6 level
DNA lesions formation
CRP level
ROS level
SSBs level
DNA repair
HAE level
DNA lesions formation
MDA level
DNA strand breaks
8-OHdG level
Normal level
Zn
Deficiency
telomeres integrity
8-oxodG level
PARP-1 activity
DDB2 expression
PARP-1 expression
RAD23B expression
DNA methylation
OGG1 expression
p53 activity
SSBs level
8-OHdG level
DNA strand breaks
CPDs formation
DNA damage accumulation
chromosome aberrations
Normal level
Vit. E
Deficiency
DNA lesions formation
ROS level
IL-6 level
8-oxodG level
BER efficiency
SSBs level
NER efficiency
DNA lesions/adducts formation
Normal level
chromosome aberrations
Vit. C
Deficiency
XRCC1 expression
CRP level
TET1 expression
8-oxodG level

Homocysteine and Genomic Stability

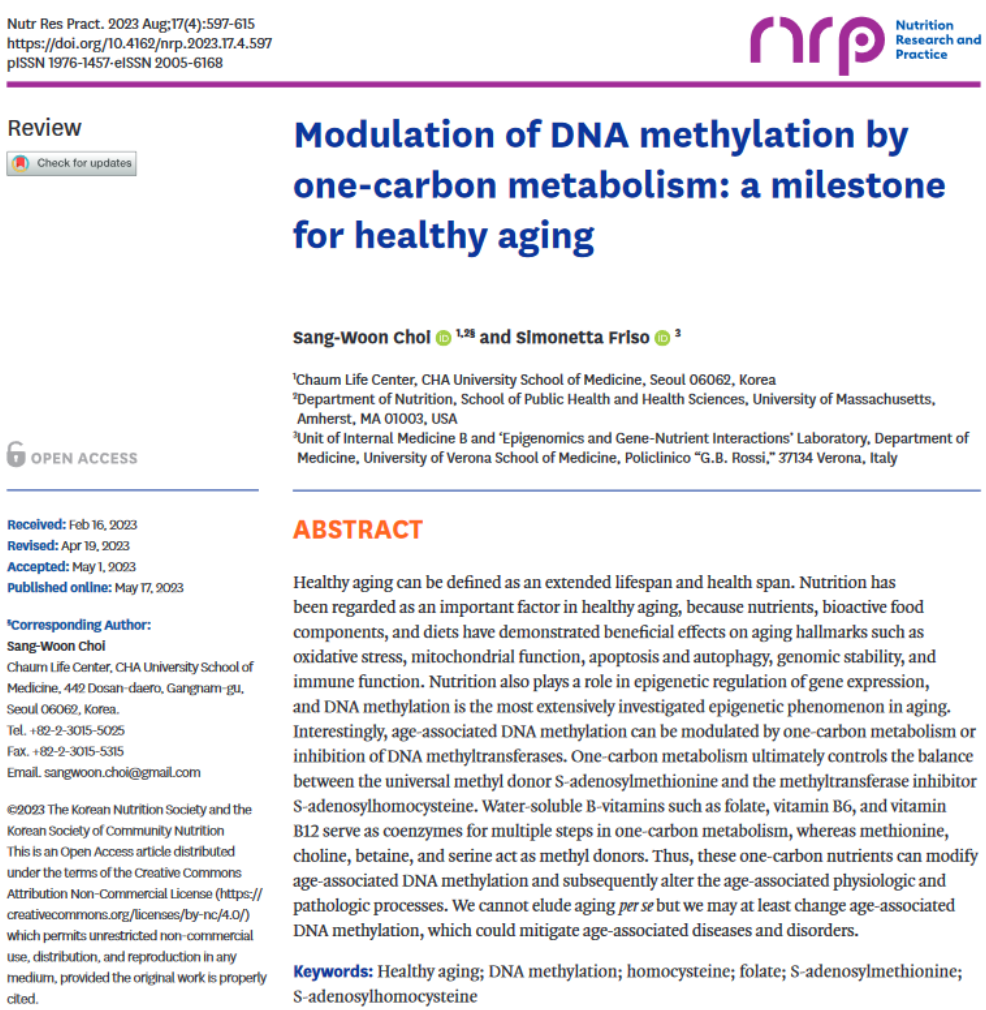
- Hyperhomocysteinemia
  - Decreased mitochondrial respiration
  - Diminshed ATP production
  - Can lead to auto-oxidation
  - Leads to ROS generation in the mitochondria



Int. J. Mol. Sci. 2020, 21, 7698

One-Carbon Metabolism

- Control balance between the universal methyl donor S-adenosylmethionine and the methyltransferase inhibitor S-adenosylhomocysteine
- Coenzymes – folate, B6, B12, B2
- Methyl donors – methionine, choline, betaine, serine
- Mitigate age-associated diseases and disorders



Nutr Res Pract. 2023 Aug;17(4):597-615
https://doi.org/10.4162/nrp.2023.17.4.597
pISSN 1976-1457-eISSN 2005-6168

Nutrition Research and Practice

Review

Check for updates

Modulation of DNA methylation by one-carbon metabolism: a milestone for healthy aging

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Email. sangwoon.choi@gmail.com

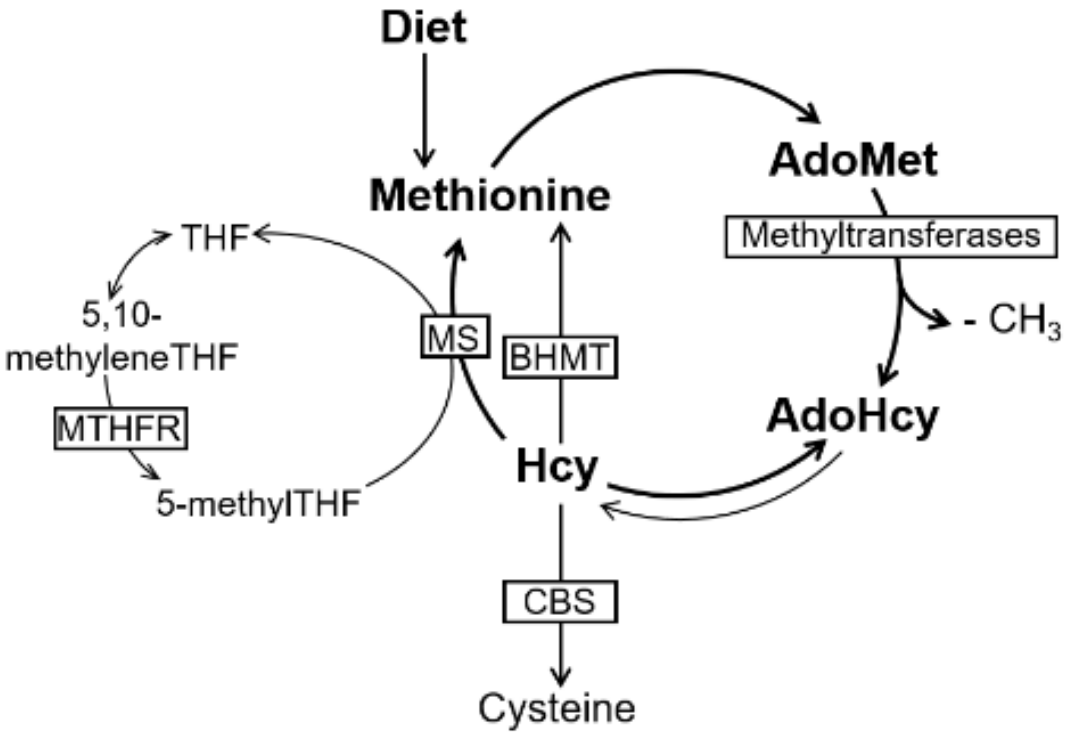
ABSTRACT

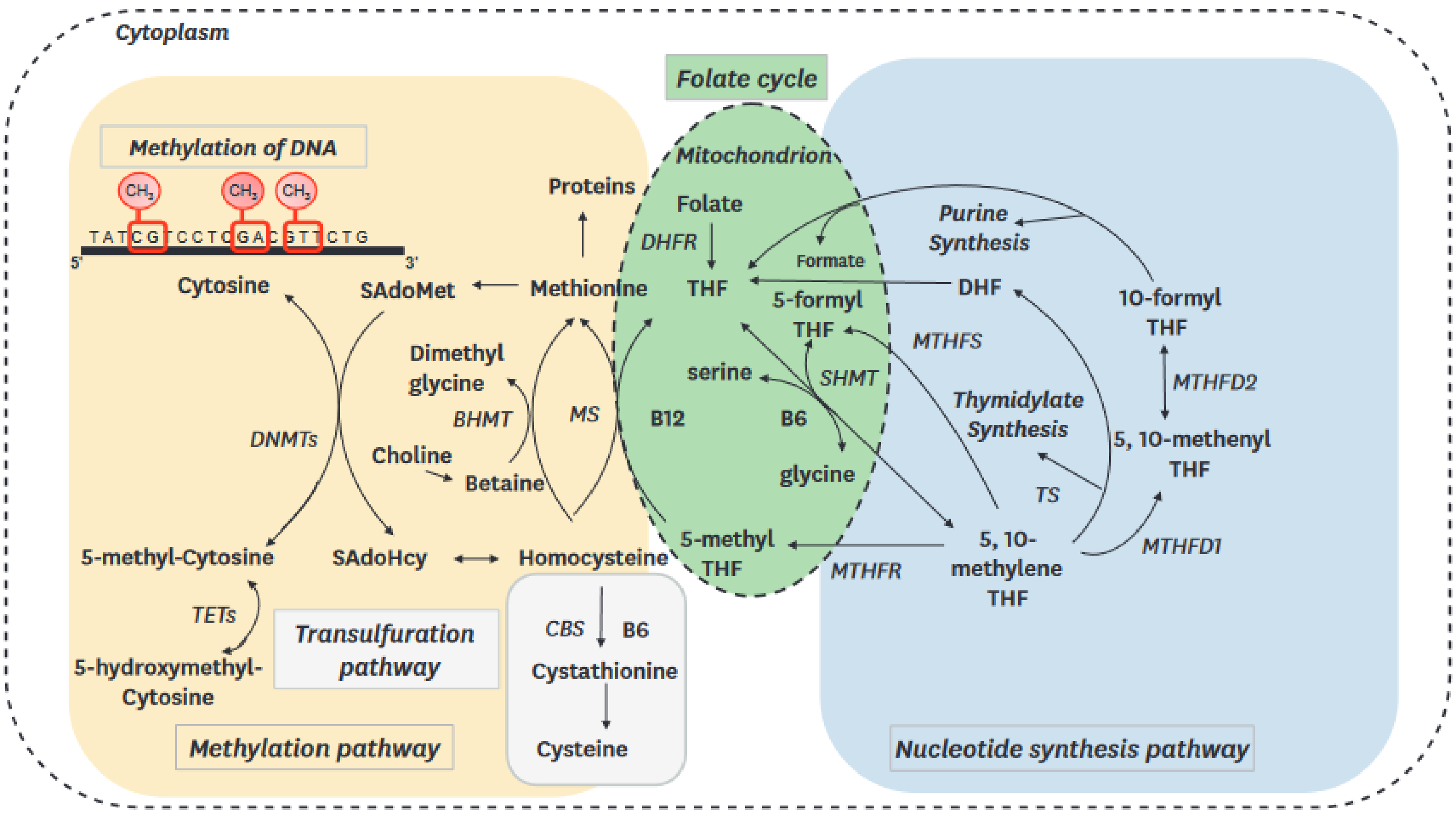
Healthy aging can be defined as an extended lifespan and health span. Nutrition has been regarded as an important factor in healthy aging, because nutrients, bioactive food components, and diets have demonstrated beneficial effects on aging hallmarks such as oxidative stress, mitochondrial function, apoptosis and autophagy, genomic stability, and immune function. Nutrition also plays a role in epigenetic regulation of gene expression, and DNA methylation is the most extensively investigated epigenetic phenomenon in aging. Interestingly, age-associated DNA methylation can be modulated by one-carbon metabolism or inhibition of DNA methyltransferases. One-carbon metabolism ultimately controls the balance between the universal methyl donor S-adenosylmethionine and the methyltransferase inhibitor S-adenosylhomocysteine. Water-soluble B-vitamins such as folate, vitamin B6, and vitamin B12 serve as coenzymes for multiple steps in one-carbon metabolism, whereas methionine, choline, betaine, and serine act as methyl donors. Thus, these one-carbon nutrients can modify age-associated DNA methylation and subsequently alter the age-associated physiologic and pathologic processes. We cannot elude aging *per se* but we may at least change age-associated DNA methylation, which could mitigate age-associated diseases and disorders.

Keywords: Healthy aging; DNA methylation; homocysteine; folate; S-adenosylmethionine; S-adenosylhomocysteine

Folate, Methionine and Transulfuration

- Cytoplasm, nucleus and mitochondria
- One-carbon metabolism supports methylation of;
  - DNA
  - Proteins including histones
  - Amino acids
  - Lipids
  - Neurotransmitters and small molecules
- Controls
  - Gene expression
  - Protein function
  - Healing processes
  - Cell energy
  - Neurological function
  - Liver detoxification
  - Immune function

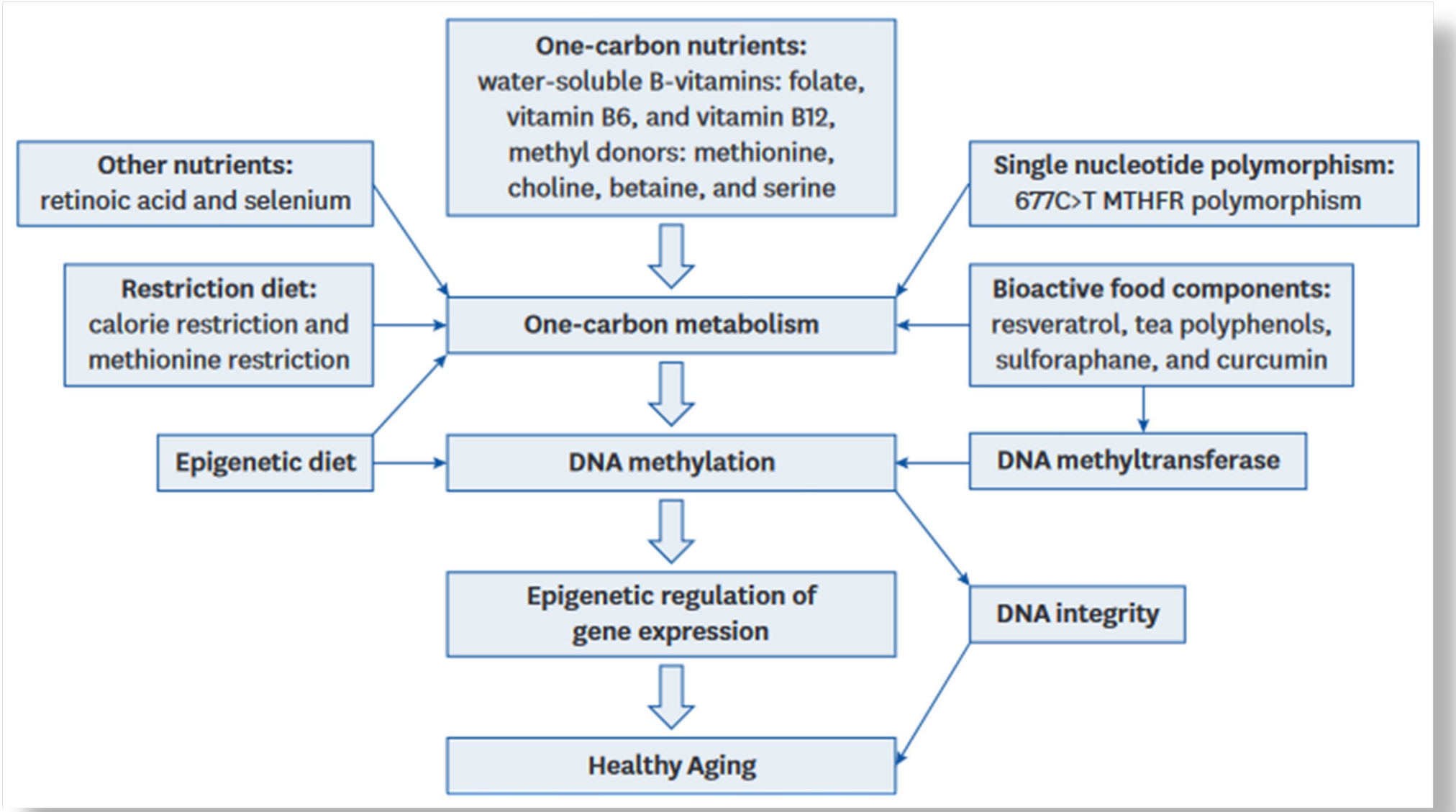



Cytoplasm
Folate cycle
Methylation of DNA
CH3
CH3
CH3
TATCGTCCTCGACGTTCTG
5'
3'
Mitochondrion
Proteins
Folate
DHFR
Formate
Purine Synthesis
Cytosine
SAdoMet
Methionine
THF
DHF
5-formyl THF
10-formyl THF
MTHFS
Dimethyl glycine
serine
SHMT
MTHFD2
BHMT
MS
B12
B6
Thymidylate Synthesis
5, 10-methenyl THF
DNMTs
Choline
Betaine
glycine
TS
5-methyl-Cytosine
SAdoHcy
Homocysteine
5-methyl THF
MTHFR
5, 10-methylene THF
MTHFD1
TETs
5-hydroxymethyl-Cytosine
Transulfuration pathway
CBS
B6
Cystathionine
Cysteine
Methylation pathway
Nucleotide synthesis pathway

Aging and Hyperhomocysteinemia

- Advancement in age tends to induce hyperhomocysteinemia
- Associated with decreased renal excretion of homocysteine
- Plasma levels increase
- Reenters the cell, conversion to S-adenosylhomocysteine, possible inhibition of methyl transfer to DNA
- Inactivation of nitric oxide synthase pathway

Comprehensive Intervention



Telomeres and B12

- Telomeres
  - Non-coding
  - Confer chromosomal stability
  - Protect the genome from nucleolytic degradation
  - Avoid aberrant recombination and improper repair
  - Prevent random fusion of chromosomes
  - Shorten with every cell division
  - Biological age indicator
  - Possess guanine rich repeats (highly susceptible to oxidative stress)
  - B12 deficiency = hyperhomocysteinemia = compromised telomere

Vitamin D and Telomeres

Vitamin D

- Cellular differentiation
- Proliferation
- Apoptosis

Higher vitamin D

- Longer telomere length
- Functions through anti-inflammatory and anti-cell proliferation mechanisms

Short telomere signals exit from the cell cycle (senescence)

J Frailty Aging. 2021;10(1):2-9.

Telomere Nutrient Relationships

- Vitamin D
- Folate
- B12
- Magnesium
  - Telomerase enzyme

- Vitamin D
  - < 30 overt deficiency
  - 30-60 – suboptimal
  - >60 – optimal
- Homocysteine
  - < 7 optimal
  - 7-9 – B12
  - >9 – Folate

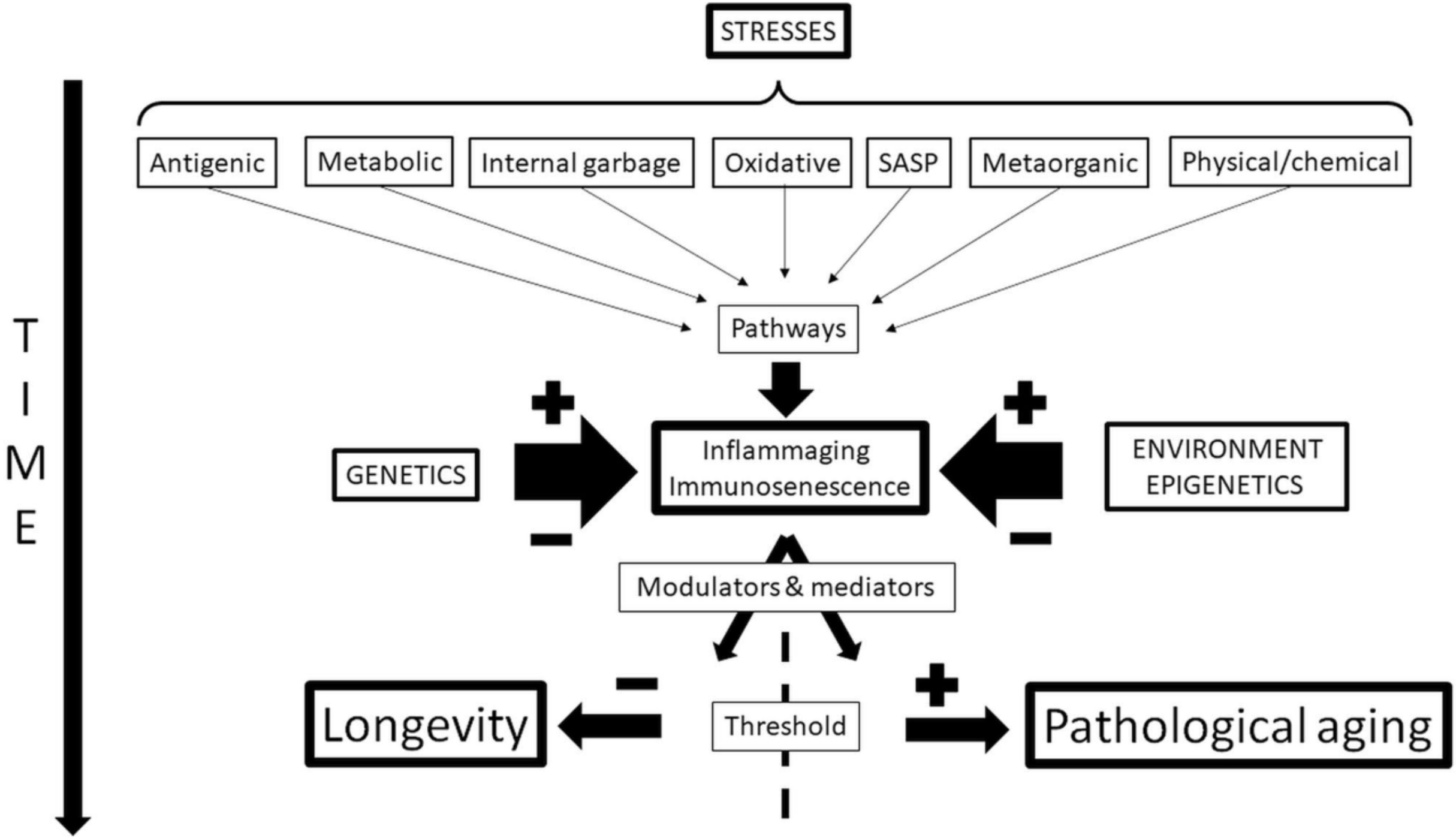
Inflammaging

- Immune changes with aging
- Adaptive immunity
- Thymic involution
  - Starts early in life
  - Complete shrinkage of the T cell priming tissue
  - Decreased naïve T cells
  - Repeat virus reactivation
- Pro-inflammatory cytokines are produced
- Stimulate other cells to become metabolically active, but immunological non-functional

Inflammaging & “Garb-aging”

- Immune system reflects the lifelong accumulation of stressors
- Macrophage centered
- Multi-system
- Includes gut microbiota
- Innate immune system recognizes
  - Endogenous / self, misplaced, altered molecules
  - Results from damaged and / or dead cells and organelles
- Decline in autophagy / mitophagy
- Sets the stage for autoreactive / autoimmune processes

Trends Endocrinol Metab. 2017 Mar;28(3):199-212.


STRESSES
Antigenic
Metabolic
Internal garbage
Oxidative
SASP
Metaorganic
Physical/chemical
Pathways
TIME
GENETICS
Inflammaging
Immunosenescence
ENVIRONMENT
EPIGENETICS
Modulators & mediators
Longevity
Threshold
Pathological aging

T cell control of inflammaging

T cells (adaptive immune system) might be the immune population most affected by aging

Directly kill infected or transformed cells

Amplify immune response

Produce cytokines

Tightly regulated

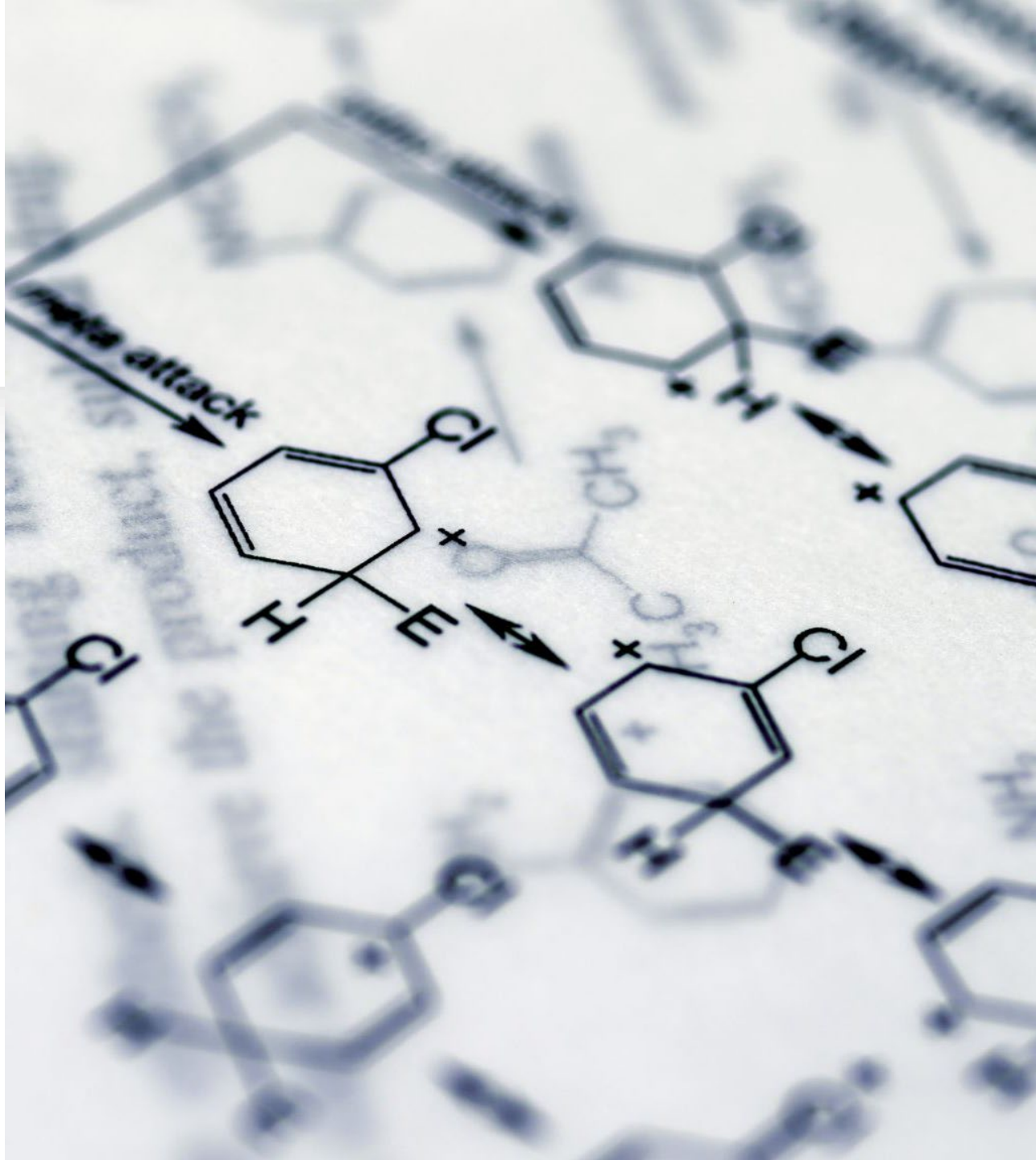
Age-related changes can significantly change the T cell landscape and function

- Reduced immune surveillance
- Onset and disease progression

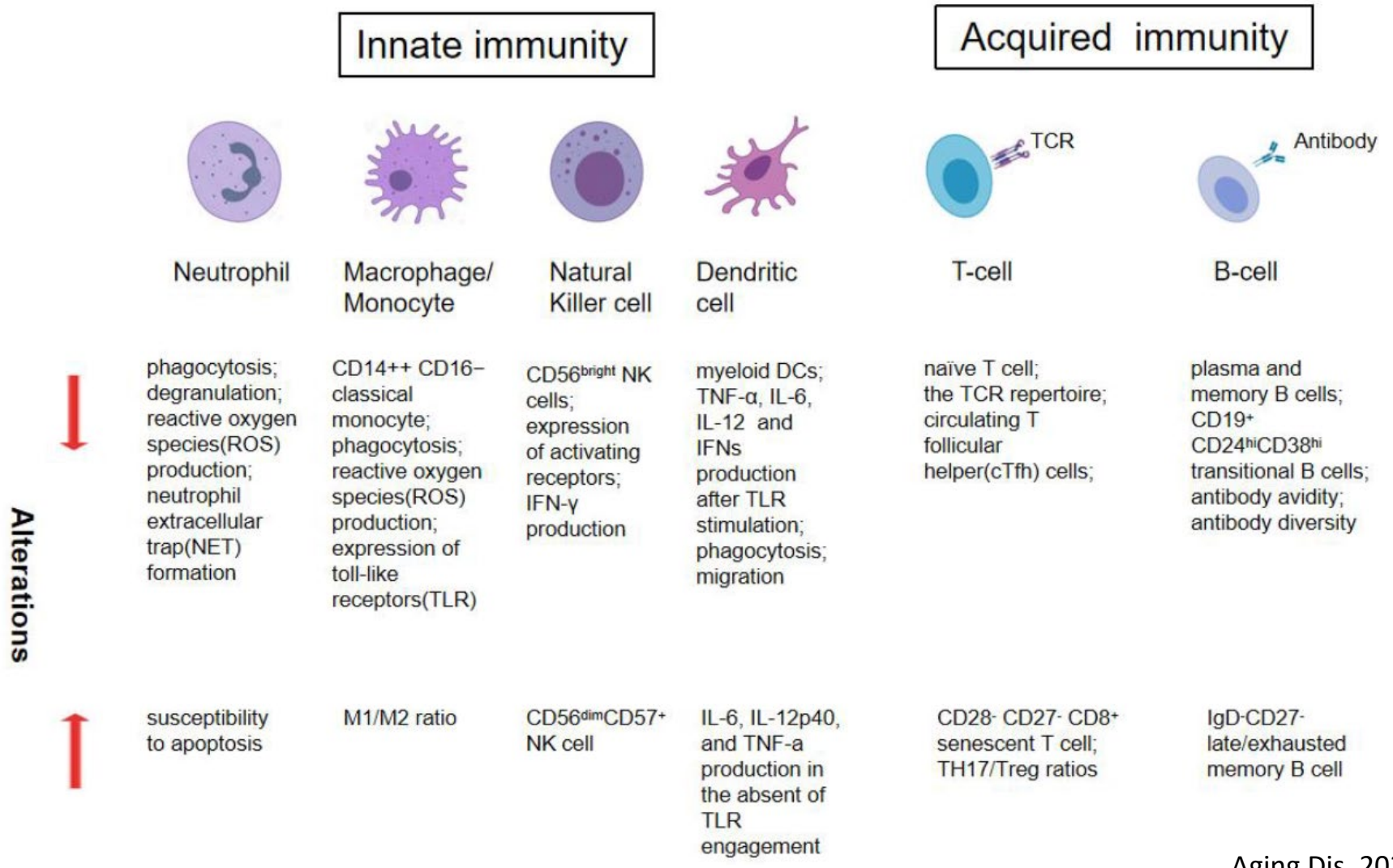
Seminars in Immunology 70 (2023) 101818

Inflammaging and Labs

- Lab indicators of inflammatory mediators
  - IL-6, TNFα, IL-1, and CRP
- Complete Blood Count
  - Neutrophils, lymphocytes, monocytes

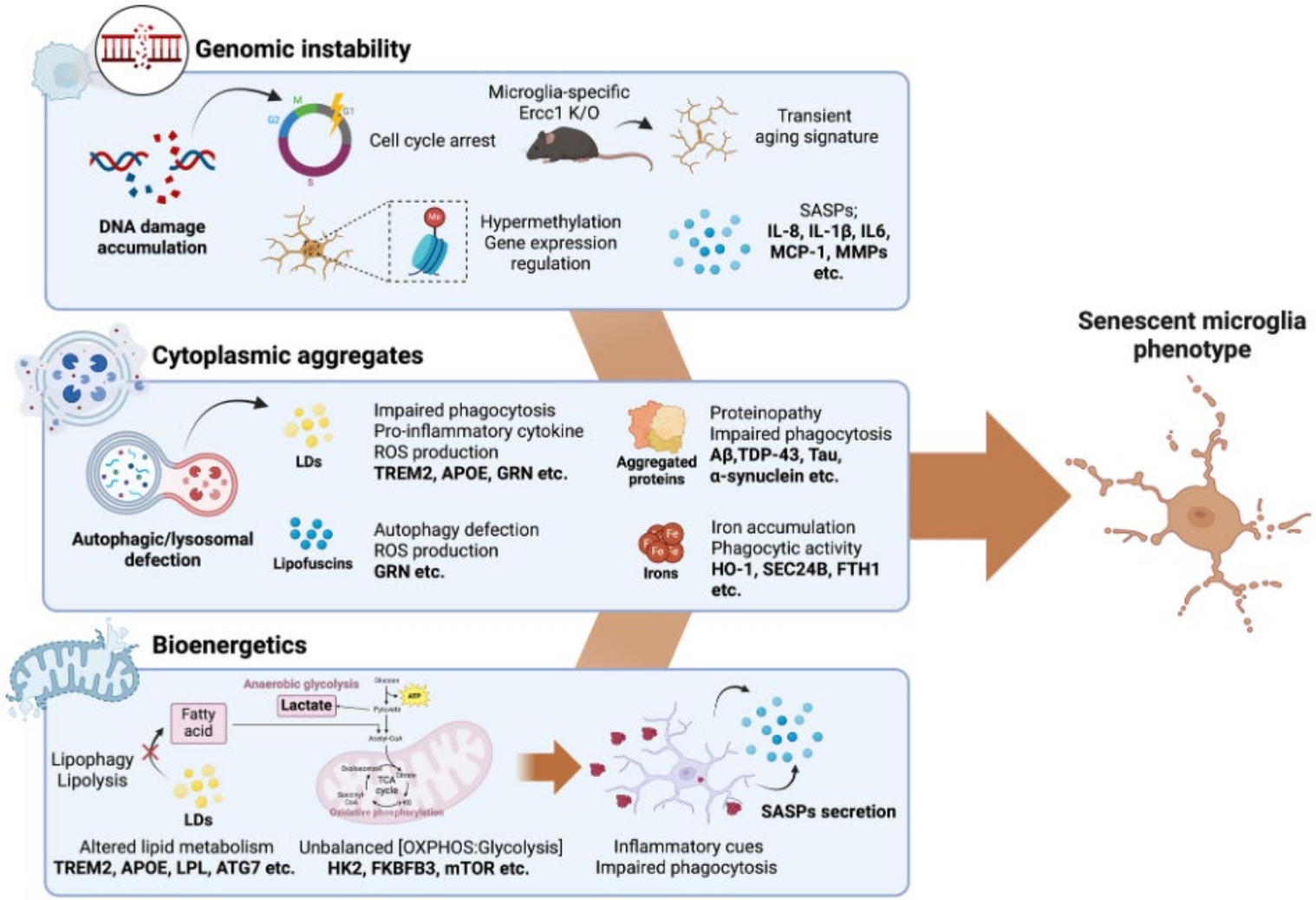


Immune Cell Alterations with Aging



Aging Dis. 2024 Aug 1;15(4):1588-1601.

Inflammaging Example



REVIEW Open Access

Emerging role of senescent microglia in brain aging-related neurodegenerative diseases

Chan Rim[1†], Min-Jung You[1†], Minyeop Nahm[3] and Min-Soo Kwon[1,2*]

Abstract

Brain aging is a recognized risk factor for neurodegenerative diseases like Alzheimer's disease, Parkinson's disease, and amyotrophic lateral sclerosis (ALS, Lou Gehrig's disease), but the intricate interplay between brain aging and the pathogenesis of these conditions remains inadequately understood. Cellular senescence is considered to contribute to cellular dysfunction and inflammaging. According to the threshold theory of senescent cell accumulation, the vulnerability to neurodegenerative diseases is associated with the rates of senescent cell generation and clearance within the brain. Given the role of microglia in eliminating senescent cells, the accumulation of senescent microglia may lead to the acceleration of brain aging, contributing to inflammaging and increased vulnerability to neurodegenerative diseases. In this review, we propose the idea that the senescence of microglia, which is notably vulnerable to aging, could potentially serve as a central catalyst in the progression of neurodegenerative diseases. The senescent microglia are emerging as a promising target for mitigating neurodegenerative diseases.

Keywords Senescent microglia, Rejuvenation, Neurodegenerative diseases, Brain aging

Introduction

It is clear that addressing 'aging' presents an appealing approach to treating neurodegenerative diseases [1]. Advances in computer and biological sciences, particularly in next-generation sequencing technologies and machine learning, have facilitated the establishment of cell-type-specific aging clocks. These clocks utilize transcriptomic and phenotypical data from specific cells to predict biological aging [2]. In alignment with the aging clocks, the Mouse Aging Cell Atlas, also known as the Tabula Muris Senis, has been developed at a single-cell resolution to comprehend the cellular characteristics of the entire mouse organ [3]. Human plasma proteome analysis has revealed organ-specific aging differences using machine learning models, indicating that most human aging may be initiated by acceleration in a single organ [4]. Particularly, accelerated brain and vascular aging predict Alzheimer's disease (AD) progression [4]. Although previous studies have provided valuable insights into the cellular landscape and molecular changes associated with aging in mice, our understanding of the cellular functions and metabolism contributing to brain aging phenotypes remains limited. Recent studies propose that senescent microglia, rather than reactive microglia, could be a novel therapeutic target in neurodegenerative diseases [5–7]. In this paper, we

†Chan Rim and Min-Jung You contributed equally to this review

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[2] Brainimmunex Inc., 26 Yatap-ro, Bundang-gu, Seongnam-si, Gyeonggi-do 13522, Republic of Korea
[3] Dementia Research Group, Korea Brain Research Institute, Daegu, Republic of Korea

Proteostasis vs. Proteinopathies

Proteostasis

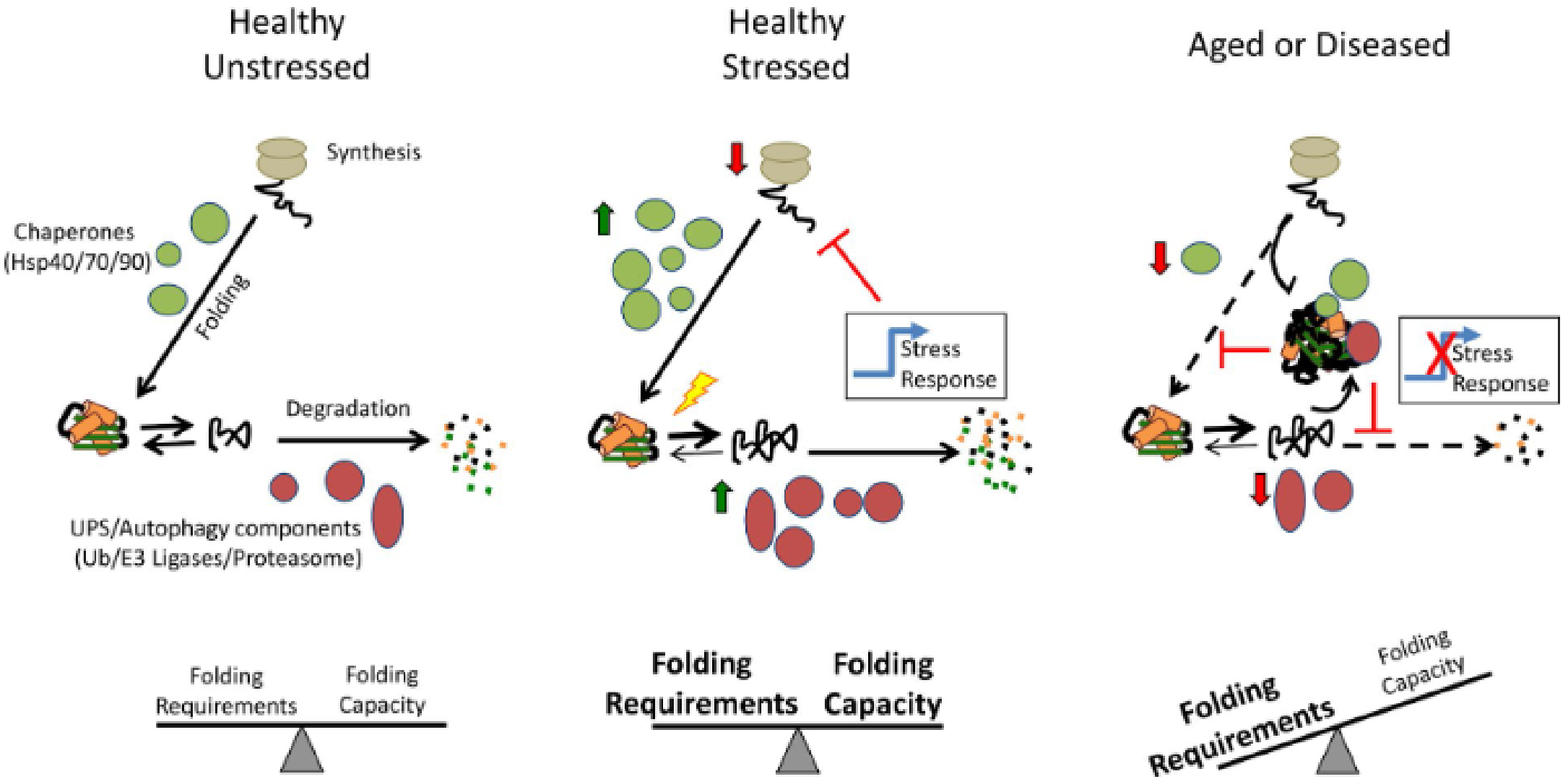
State of balanced processes of protein translation, folding, maintenance, and degradation.

Balanced neurological and bioenergetic function

Proteinopathies

- Accumulation of misfolded proteins
- Generation of dysfunctional proteins
- Aging brain
- Tauopathies, α-synucleinopathies

Translational Neurodegeneration (2024) 13:10



J. Cell Biol. Vol. 217 No. 1 51–63

Successful Aging Defined

Extended, healthy life

Absence of serious, debilitating, painful age-related diseases

Reduced focus on longevity

Enhanced focus on healthspan and quality of life

Inflammaging and Cannabinoids

- Immuno-modulatory
- CBD (cannabidiol) and CBC (cannabichromene)
- Inflammatory response regulation
- Normalization of circadian rhythm (biomarker for aging)
- Suppression of oxidative stress (antioxidants)
- Anxiolytic properties
- Repression of ROS / RNS

Microbiome and Inflammaging

- Alteration in microbiome composition, function, metabolic output, phenotype and diversity impact immune balance and homeostasis
- Senescence-induced dysfunction affects the tolerogenic and symbiotic relationship between microbiome and Immune system
- Cannabinoids re-establish balance and symbiosis between microbiota and immune system
  - Specifically, CBD
- Optimizes endocannabinoid system, regulating intestinal permeability
- Reduced progression towards autoimmunity

Ageing Res Rev. 2021 December ; 72: 101487.

Aging with Rhythmicity

ELSEVIER

Life Sciences

journal homepage: www.elsevier.com/locate/lifescie

Review article

Aging with rhythmicity. Is it possible? Physical exercise as a pacemaker

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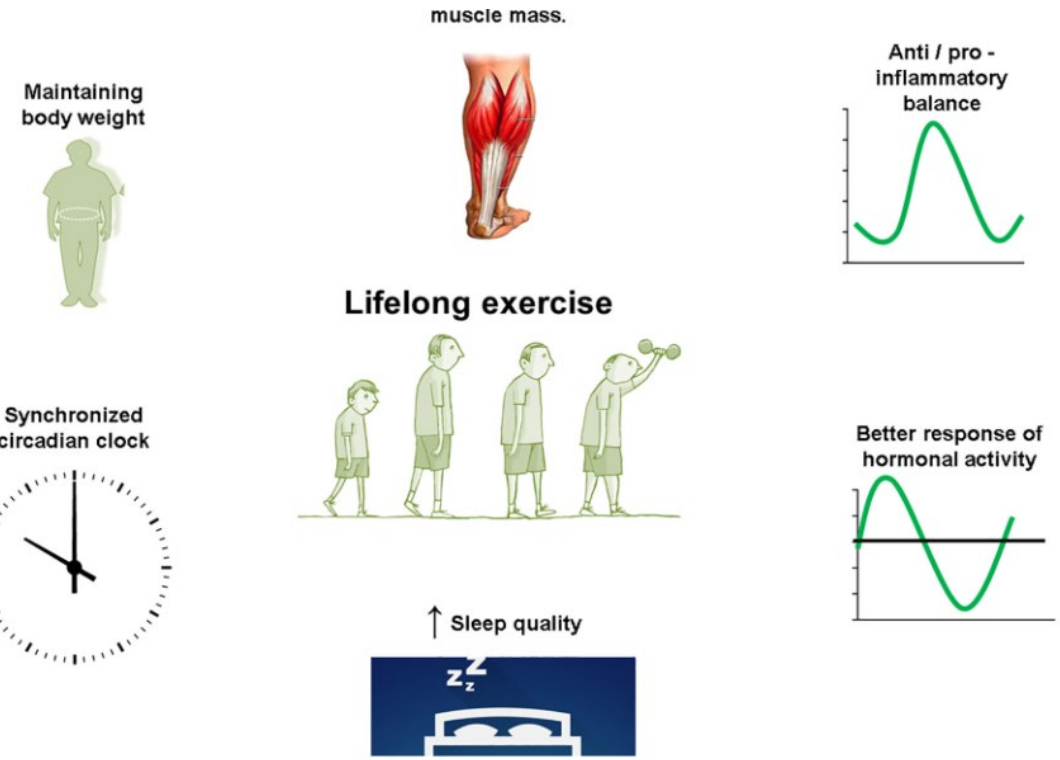
ARTICLE INFO

Keywords:
Aging
Inflammaging
Immunosenescence
Circadian rhythms
Clock genes

ABSTRACT

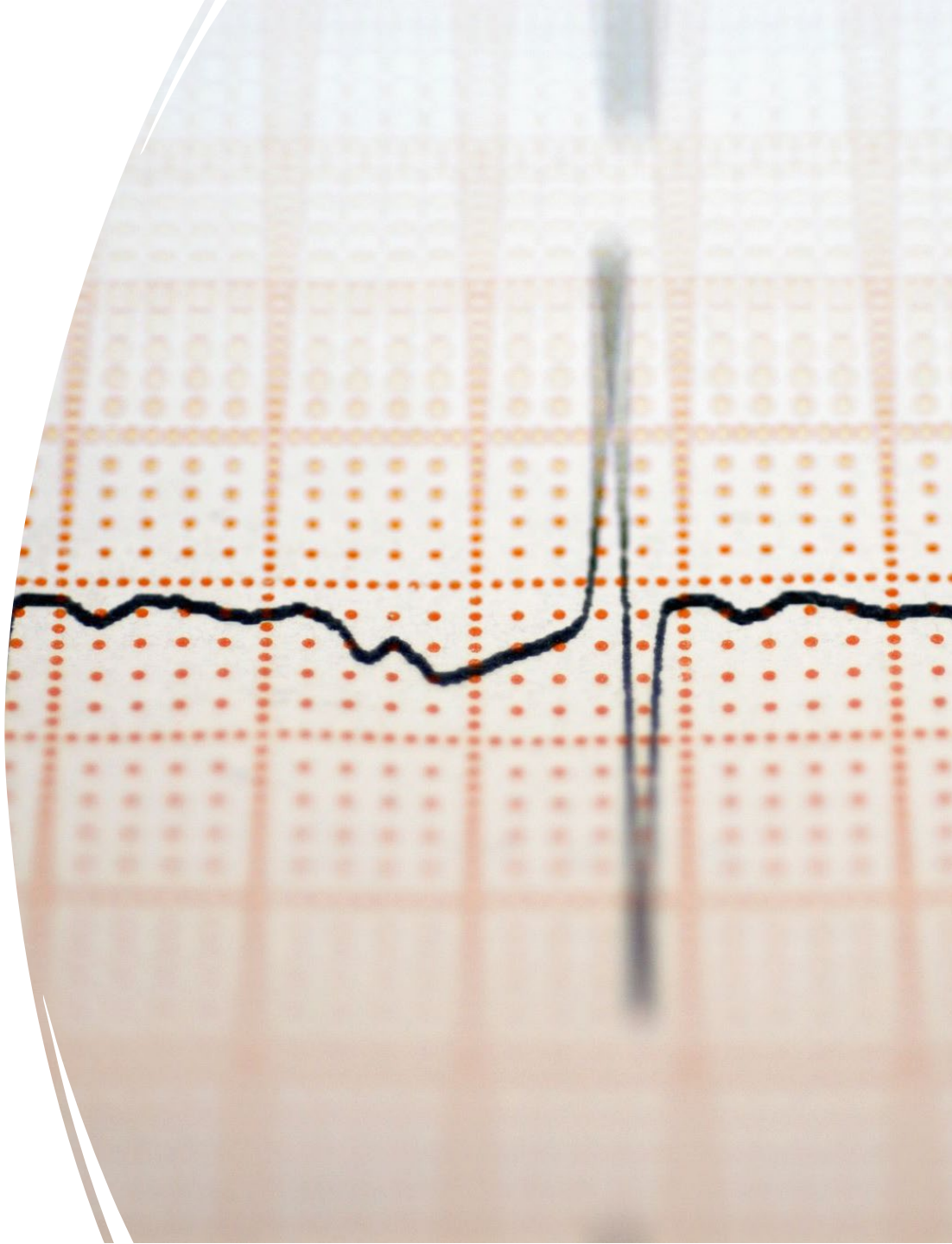
Aging is associated with gradual decline in numerous physiological processes, including a reduction in metabolic functions and immunological system. The circadian rhythm plays a vital role in health, and prolonged clock disruptions are associated with chronic diseases. The relationships between clock genes, aging, and immunosenescence are not well understood. Inflammation is an immune response triggered in living organisms in response to the danger associated with pathogens and injury. The term 'inflammaging' has been used to describe the chronic low-grade-inflammation that develops with advancing age and predicts susceptibility to age-related pathologies. Equilibrium between pro-and anti-inflammatory cytokines is needed for healthy aging and longevity. Sedentary and poor nutrition style life indices a disruption in circadian rhythm promoting an increase in pro-inflammatory factors or leads for chronic low-grade inflammation. Moreover, signals mediated by pro-inflammatory cytokines, such as tumor necrosis factor-alpha and interleukin-6, might accentuate of the muscle loss during aging. Circadian clock is important to maintain the physiological functions, as maintenance of immune system. A strategy for imposes rhythmicity in the physiological systems may be adopted of exercise training routine. The lifelong regular practice of physical exercise decelerates the processes of aging, providing better quality and prolongation of life. Thus, in this review, we will focus on how aging affects circadian rhythms and its relationship to inflammatory processes (inflammaging), as well as the role of physical exercise as a regulator of the circadian rhythm, promoting aging with rhythmicity.



Cardiovascular Aging and Intervention

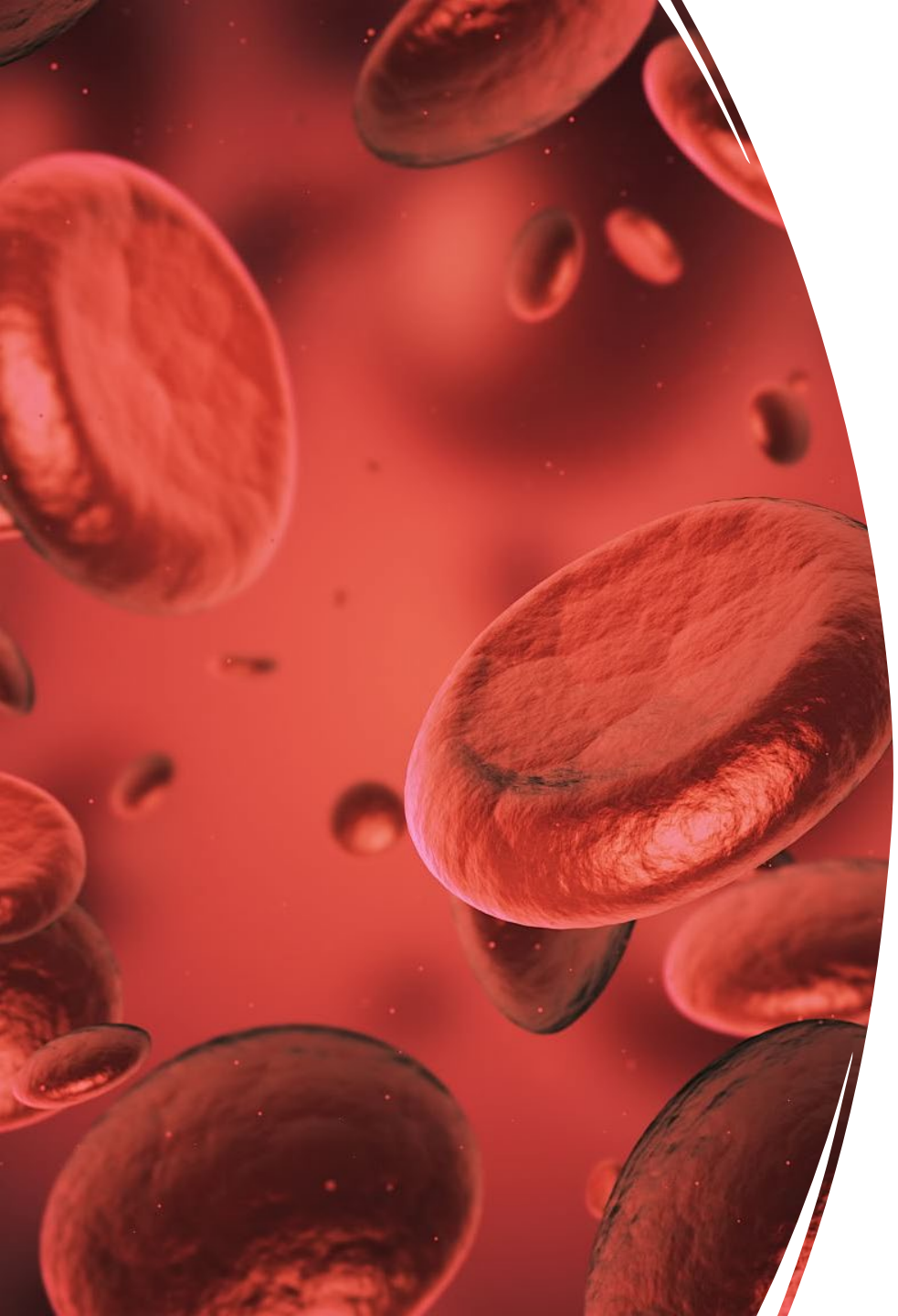
- Gender differences
  - Females
    - Loss of estrogen activates inflammatory pathways
    - Higher levels of inflammatory cytokines
    - Myocardium macrophage infiltration
      - M1 macrophage (inflammatory) vs. M2 macrophage (anti-inflammatory)
  - Males
    - More inflammatory responses in myocardial tissues
    - Increased risk of cardiomyopathy and end-stage myocarditis

Translational Aspects. Cells 2022, 11, 1010.



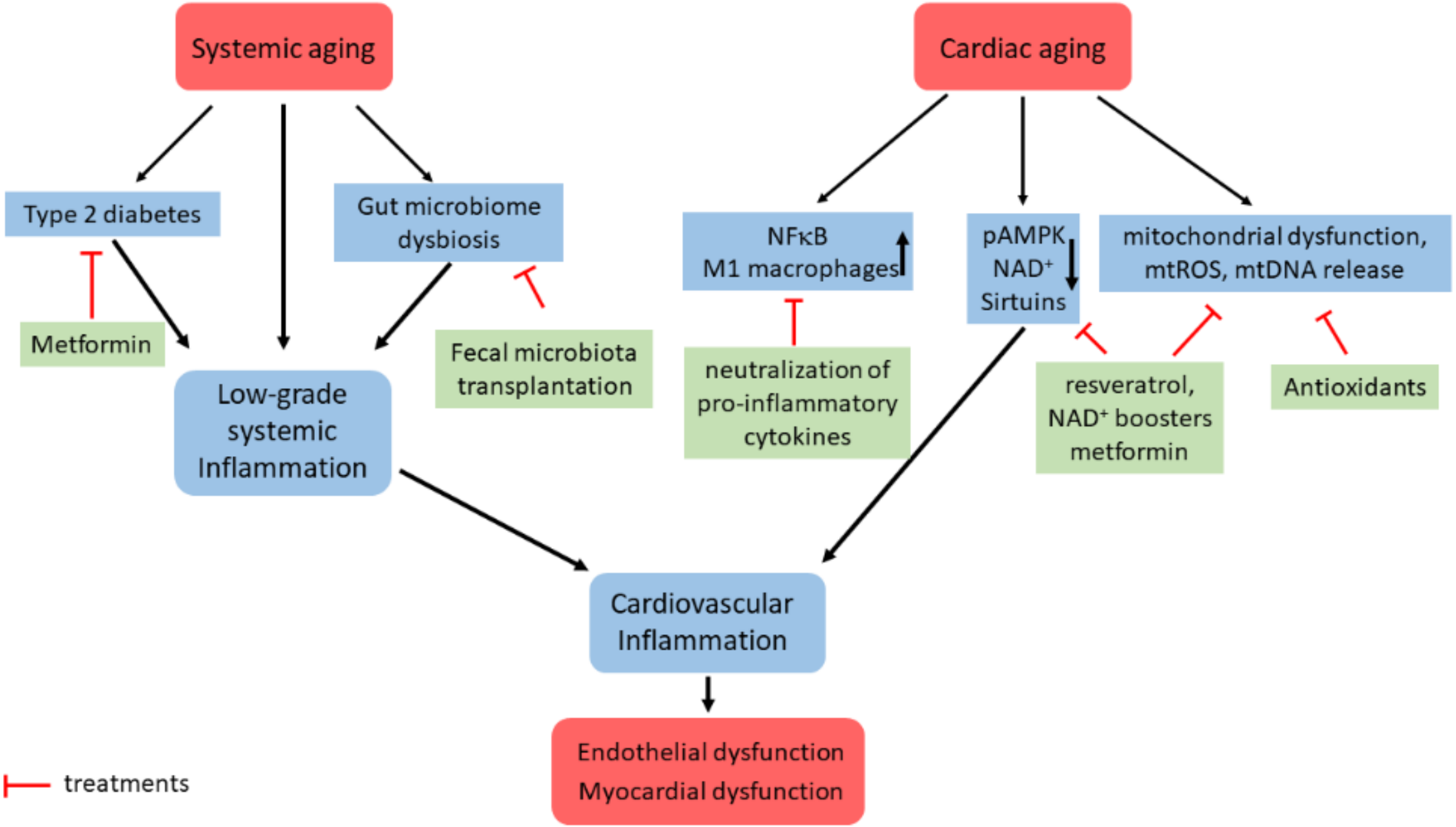
Cardiovascular Aging and Intervention

- Cardiomyocytes are high-energy consuming cells
  - Mitochondria dense
- AMPK – key energy sensor and regulator of energy metabolism and mitochondrial homeostasis
  - Activated by ATP depletion
  - Activation decreases with age
- Sirt1 (sirtuins)
  - Promotes AMPK activity
- Reduced cellular energy balance
- Decreased mitochondrial function, biogenesis and mitophagy



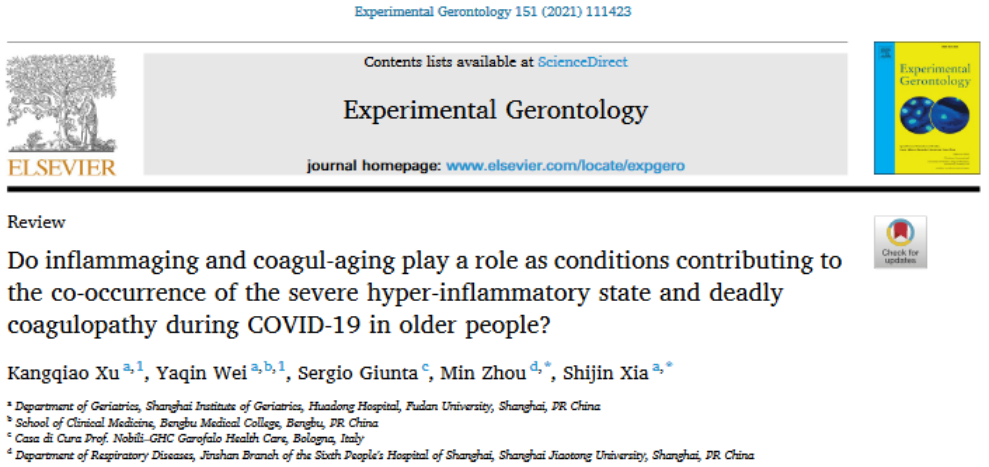
Vascular Inflammaging

- Associated with severe atherosclerosis and microvascular dysfunction
- Pathological vascular remodeling and vascular stiffness
- Can start as early as childhood
- Gradual changes in vascular structure
- Reduced vascular compliance
- Nitric oxide is released to counter
  - NO bioavailability decreases with time
- Aging causes reduced NO bioavailability


Systemic aging
Cardiac aging
Type 2 diabetes
Gut microbiome dysbiosis
NFκB
M1 macrophages
pAMPK
NAD+
Sirtuins
mitochondrial dysfunction, mtROS, mtDNA release
Metformin
Fecal microbiota transplantation
neutralization of pro-inflammatory cytokines
resveratrol, NAD+ boosters metformin
Antioxidants
Low-grade systemic Inflammation
Cardiovascular Inflammation
Endothelial dysfunction
Myocardial dysfunction
treatments

Coagul-aging

- Inflammaging
  - Change in cytokine profiles
- COVID-19
  - Cytokine storm / hyperinflammation
  - Innate immune system activation
  - Excessive inflammation leads to abnormal coagulation
  - Crosstalk
  - Physiological aging – increased blood coagulation proteins in plasma, fibrinolysis impairment
    - Exacerbated by Inflammaging



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Experimental Gerontology

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Review

Do inflammaging and coagul-aging play a role as conditions contributing to the co-occurrence of the severe hyper-inflammatory state and deadly coagulopathy during COVID-19 in older people?

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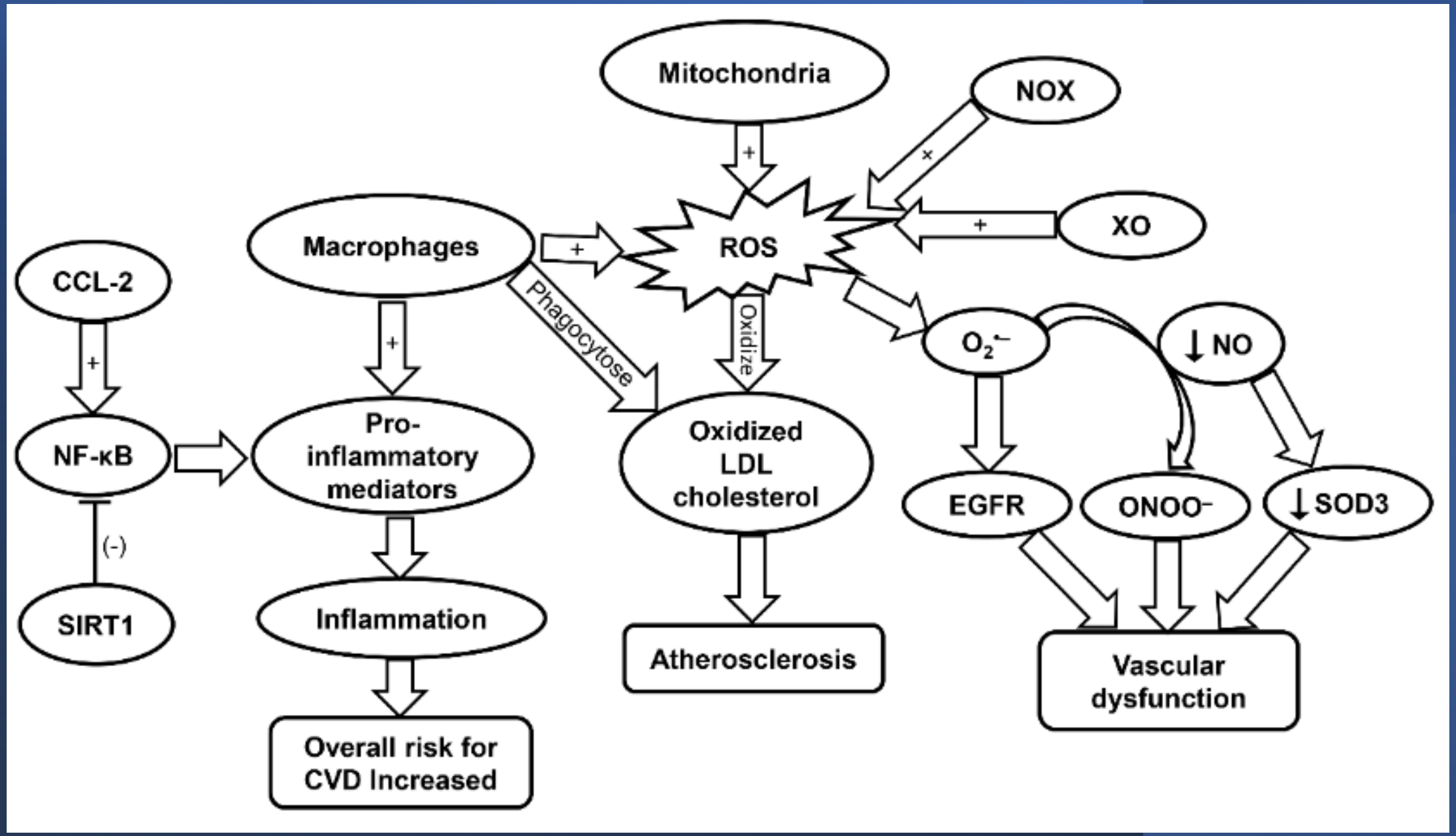
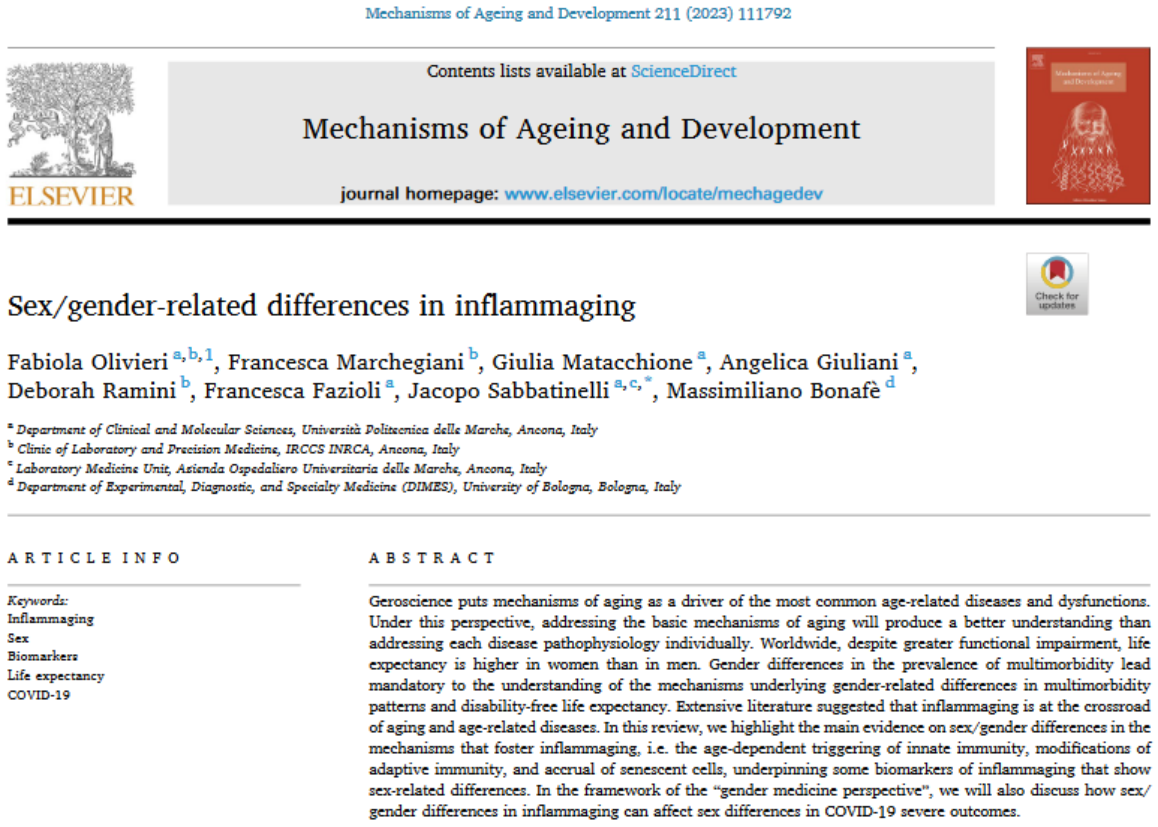

Mitochondria
NOX
+
+
ROS
+
XO
Macrophages
+
CCL-2
Phagocytose
Oxidize
$O_2^{\bullet-}$
↓ NO
+
+
NF-κB
Pro-
inflammatory
mediators
Oxidized
LDL
cholesterol
EGFR
ONOO⁻
↓ SOD3
(-)
SIRT1
Inflammation
Atherosclerosis
Vascular
dysfunction
Overall risk for
CVD Increased

Gender Differences in Inflammaging

- Women
  - Longer life expectancy
  - Greater functional impairment (unhealthy longevity)
  - Age at natural menopause linked to biological age
  - Higher phagocytic activity compared to males
  - Enhanced cellular responsiveness to the LPS-mediated stimulation
  - Lower infection rates, but fourfold increase in autoimmunity
  - Higher T cell count



Mechanisms of Ageing and Development 211 (2023) 111792

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Mechanisms of Ageing and Development

journal homepage: www.elsevier.com/locate/mechagedev

Sex/gender-related differences in inflammaging

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ARTICLE INFO

Keywords:
Inflammaging
Sex
Biomarkers
Life expectancy
COVID-19

ABSTRACT

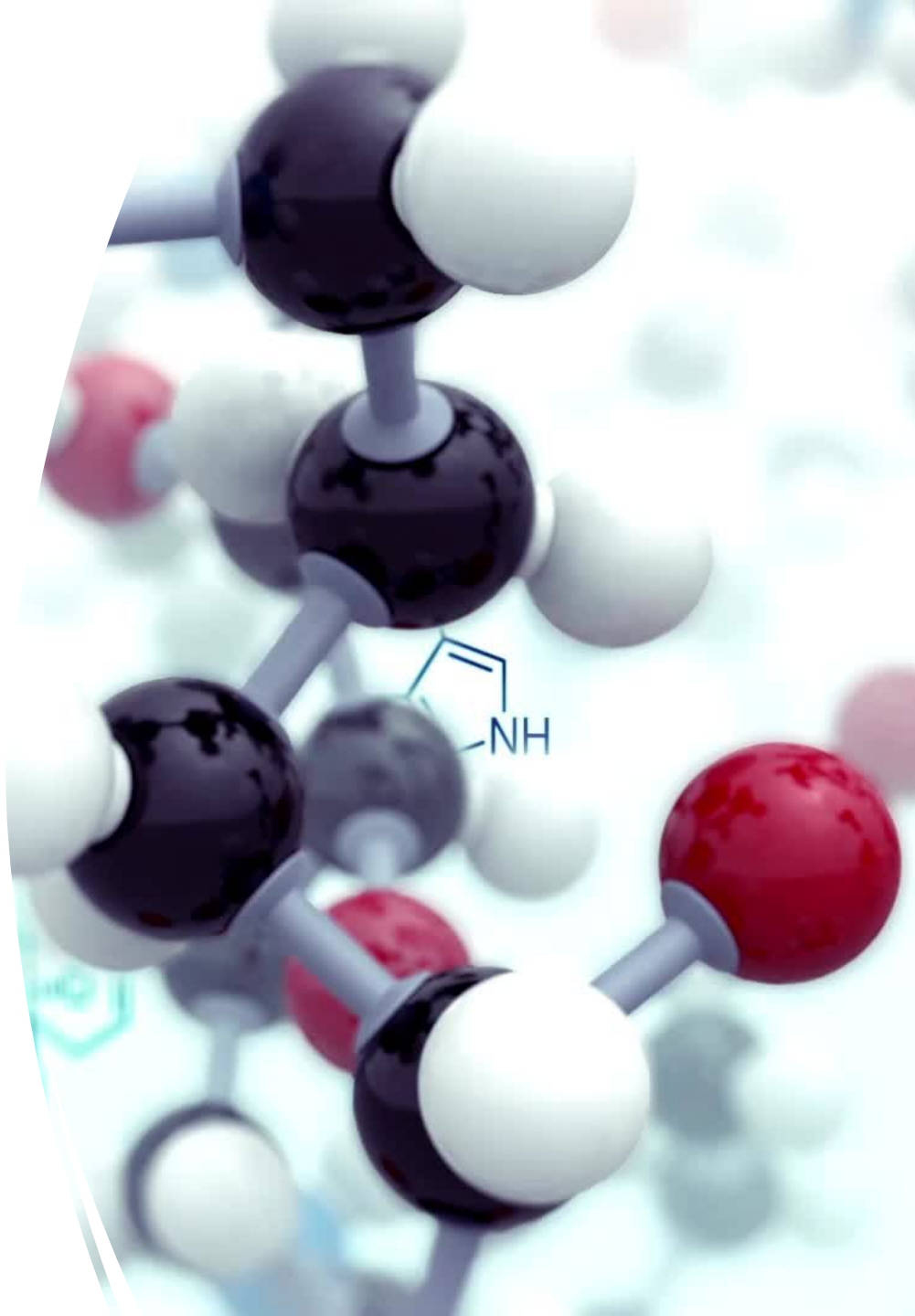
Geroscience puts mechanisms of aging as a driver of the most common age-related diseases and dysfunctions. Under this perspective, addressing the basic mechanisms of aging will produce a better understanding than addressing each disease pathophysiology individually. Worldwide, despite greater functional impairment, life expectancy is higher in women than in men. Gender differences in the prevalence of multimorbidity lead mandatory to the understanding of the mechanisms underlying gender-related differences in multimorbidity patterns and disability-free life expectancy. Extensive literature suggested that inflammaging is at the crossroad of aging and age-related diseases. In this review, we highlight the main evidence on sex/gender differences in the mechanisms that foster inflammaging, i.e. the age-dependent triggering of innate immunity, modifications of adaptive immunity, and accrual of senescent cells, underpinning some biomarkers of inflammaging that show sex-related differences. In the framework of the "gender medicine perspective", we will also discuss how sex/gender differences in inflammaging can affect sex differences in COVID-19 severe outcomes.

Oxidative Stress and Inflammaging in Human Disease

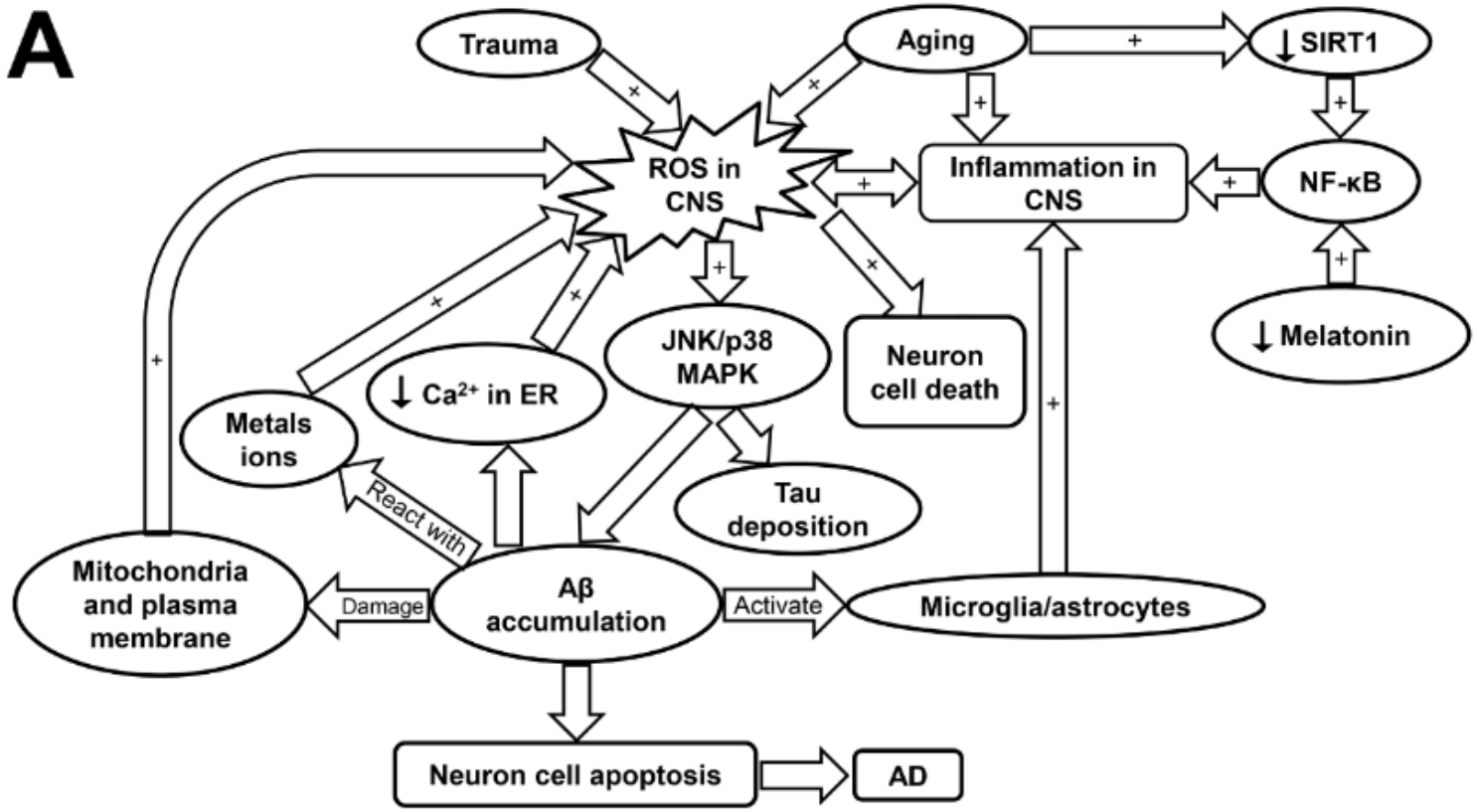
- Over-abundance of reactive oxygen species in the cell
- Leads to oxidation and damage of cellular components, increased inflammation, and activation of cell death pathways
- Cardiovascular disease, cancer, neurodegenerative diseases, chronic obstructive pulmonary disease, diabetes, and rheumatoid arthritis
- Three primary reactive oxygen species (ROS)
  - Superoxide (O2), hydrogen peroxide (H2O2), hydroxyl radical (OH)
- Necessary at low levels
  - Essential signaling molecules
- Redox balance is key
  - Disrupted with inflammation, vigorous exercise and aging

Key Antioxidants

- Glutathione peroxidase (GPX) and catalase, which both catalyze to decompose H2O2 into H2O and O2
- Superoxide dismutase (SOD), which catalyzes the conversion of O2•− into H2O2
- Non-enzymatic antioxidant defense
  - Vitamins A, C, E, melatonin, and polyphenols

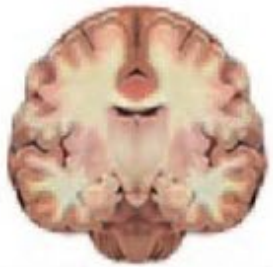


Neuro-oxidation as a Driver of Neuro-Aging



Int. J. Mol. Sci. 2019, 20, 4472

Healty brain



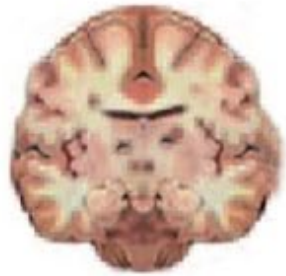
Balance between pro- and anti-inflammatory factors

Low speed decline of metabolic functions

Low intensity oxidative stress

Low level of inflammation

Aged brain



Mild cronic pro-inflammatory state

Impaired glucose and lipid metabolism

Oxidative imbalance

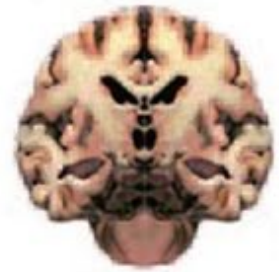
Impaired autophagic and proteasomal degradation

Impaired DNA repair

Cell senescence

Aberrant neuronal network activity

Pathological brain

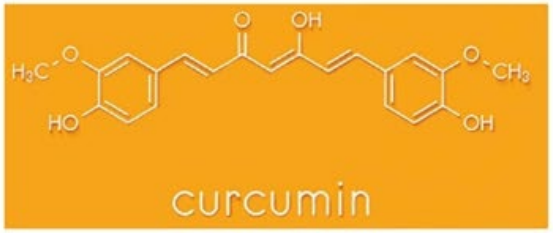


Accumulation of oxidatively damaged molecules

Extracellular deposition of proteins with altered physicochemical properties

Neuronal cell death

Exacerbated inflammatory responses



- **Antioxidant**
  - Free radicals scavenging activity
  - Antioxidant enzymes stabilisation
- **Anti inflammatory**
  - Reduction of pro-inflammatory cytokine levels
  - Inhibition of COX-2 and 5-LOX
  - Inhibition of NF-Kb
  - Inhibition of STAT3 and activation of PPAR-γ
- **Neuroprotective**
  - Activation of molecular chaperones
  - Increase of neurotrophic factors
  - Chelation of heavy metals
- **Epigenetic regulator**
  - Inhibition of HAT

Molecules 2022, 27, 341.

Atherosclerosis

↓ Oxidative stress in the aorta
Control of inflammation and senescence in the aorta

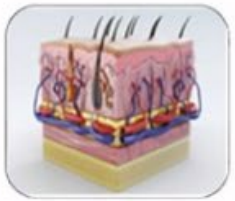
Reproductive Senescence

↑ Ovarian volume and number of follicles
Potentiated oocyte maturation and embryo development

Neurological Disorders

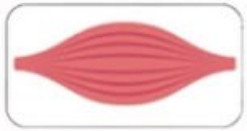
Improved motor and cognitive functions
↓ Inflammation and mitochondrial dysfunctions
↓ Oxidative stress
Neuroprotective effects

Effects of Curcumin on Aging Disorders



Skin Aging

↓ Cell death
↓ Apoptotic and inflammatory genes expression



Muscle mass loss and disfunction

Improved muscle mass and force
↓ Oxidative damage
↑ Antioxidant enzymes expression

Inflammaging and Fatty Acid Oxidation

Fatty acid oxidation – primarily β-oxidation

Fat breakdown to produce cellular energy

Relied on by myeloid cells (macrophages, microglia, monocytes)

Inflammaging damage fatty acid oxidation in myeloid cells

Excess production of AGEs (advanced glycation end products)

Increased nuclear factor-κB (NF-κB)

Decreases 5’ AMP-activated protein kinase and peroxisome proliferator-activated receptor-α

Escalates inflammation, lipid accumulation and cellular stress

Macrophage Activation

M1 (inflammatory)

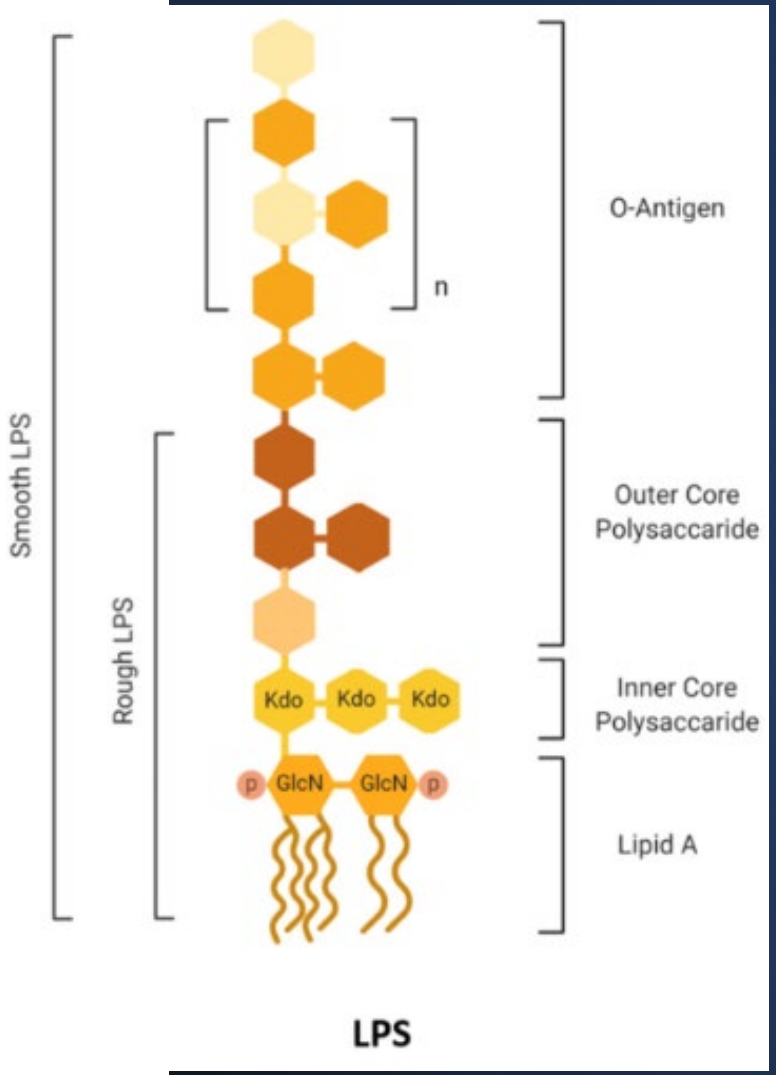
- Decreased FAO
- Increased glycolytic metabolism, nitric oxide production
- Propensity for nucleotide biosynthesis and reactive oxygen species (ROS)

M2 (anti-inflammatory)

- Increased energy demand reliant on FAO, oxidative phosphorylation, and glutamine metabolism
- Reduced utilization of glycolysis
- Uninterrupted Krebs / TCA cycle
- Aid in inflammation resolution
- Promotion of tissue homeostasis and repair

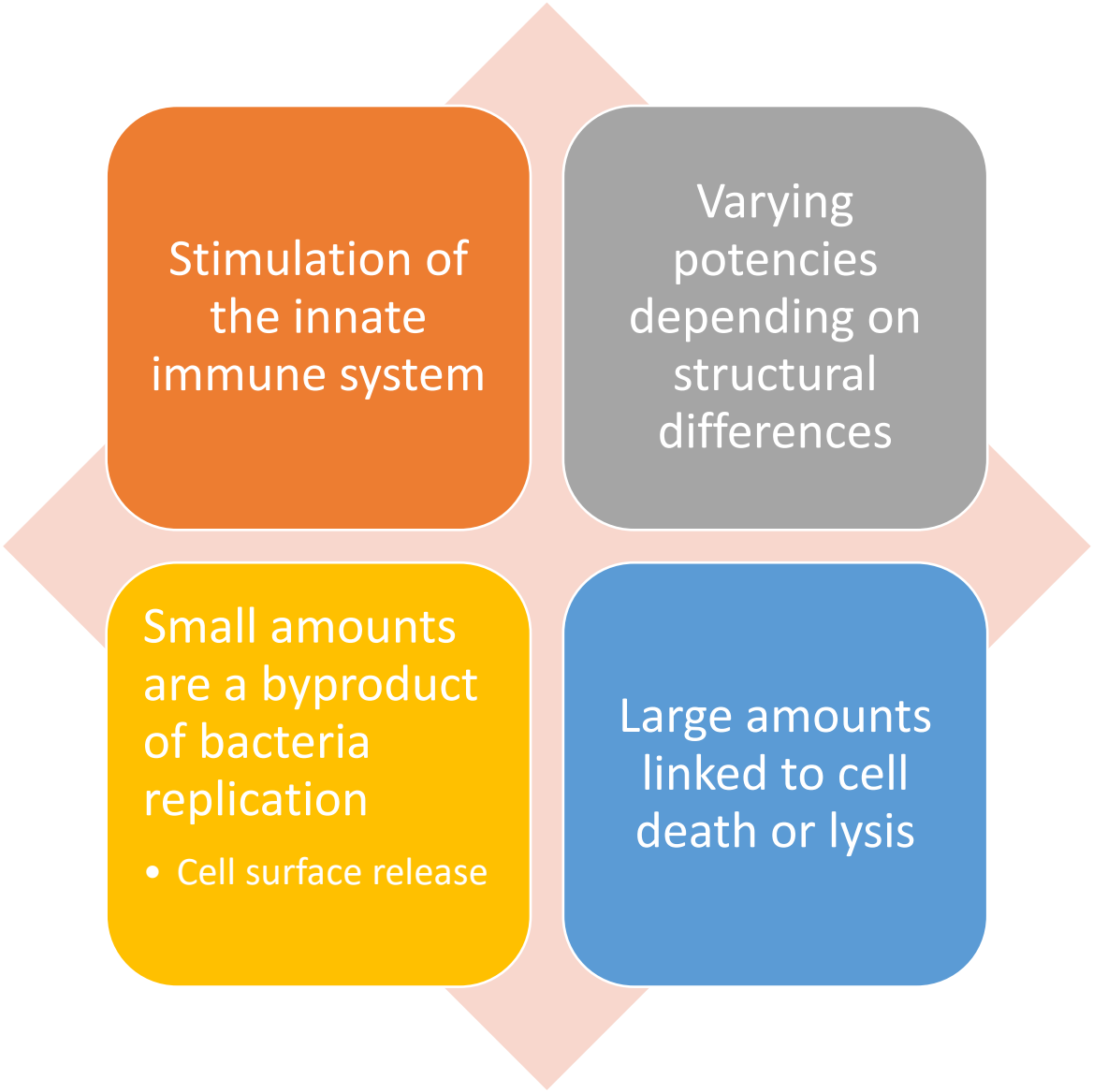
Lipopolysaccharides – Cell Signaling and Inflammation

- Lipopolysaccharides
  - Main structural component of outer membrane of most Gram-negative bacteria
  - Diverse immunostimulatory effects
  - Procoagulant activity
  - Normally present in small amounts
  - Large amounts induce immune activation
  - Leads to immune dysregulation
  - Linked to numerous progressive pathophysiological states including depression, anxiety, NAFLD, metabolic and cardiovascular disease
  - Low level, chronic endotoxemia

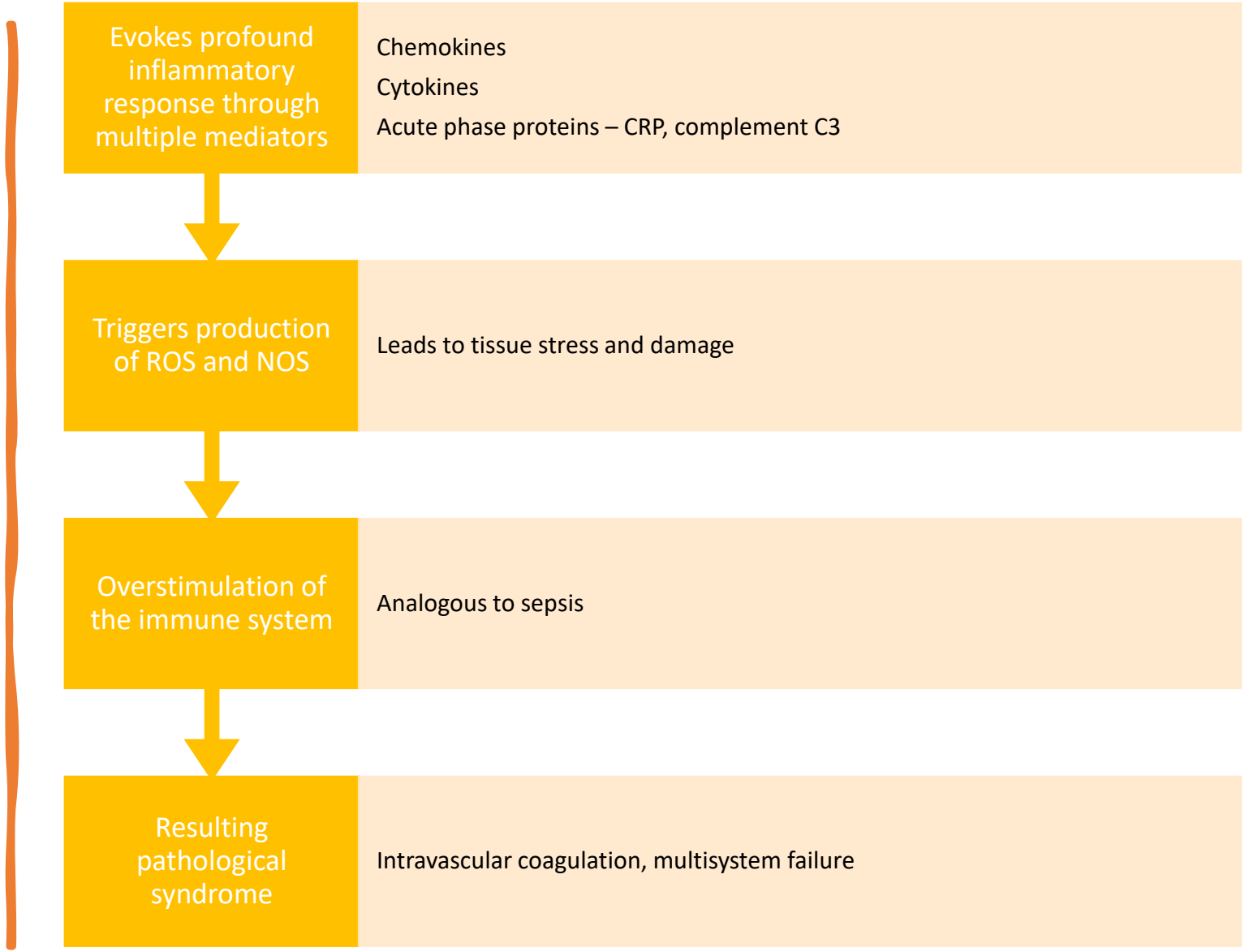


Chronic Stress
Volume 6: 1–18

Lipopolysaccharides



LPS in Chronic Disease



Interventions for LPS

Primary sites to consider

- Gut
- Oral cavity (periodontal disease)
- Catheters

Non-absorbable drugs / non-drugs that for LPS complexes

- Supplemental immunoglobulins

Strengthening the intestinal barrier

Nutritional management (improving diet quality and micronutrient status)

Manipulating gut microbiome (pre, pro and post-biotics)

Nutritional supplements

- Ginger (Zingerone)
- Anti-inflammatory antioxidants

Inflammaging and RAGE

RAGE – Receptor for Advanced Glycation End Products

Multi-ligand – multiple triggers

- 28 known
- DAMPs – damage associated molecular patterns
- PAMPs – pathogen associated molecular patterns
- Can be produced by senescent cells

Transmembrane receptor

Activates key pro-inflammatory and pro-oxidative pathways

Well noted in diabetes

Driver of physiological aging

AGING 2019, Vol. 11, No. 17

Dysbiosis

H. pylori, SIBO and More

It Starts at the Top

H. pylori

A Common Pathogen

Affects about 50% of the world's population

Severity has not significantly diminished in the last several years

Single organism

Pathogen

Highly genetically diverse

Environmentally adaptable (antibiotic resistance)

Infection

Colonizes stomach and changes gastric secretions

Pathogen Activity

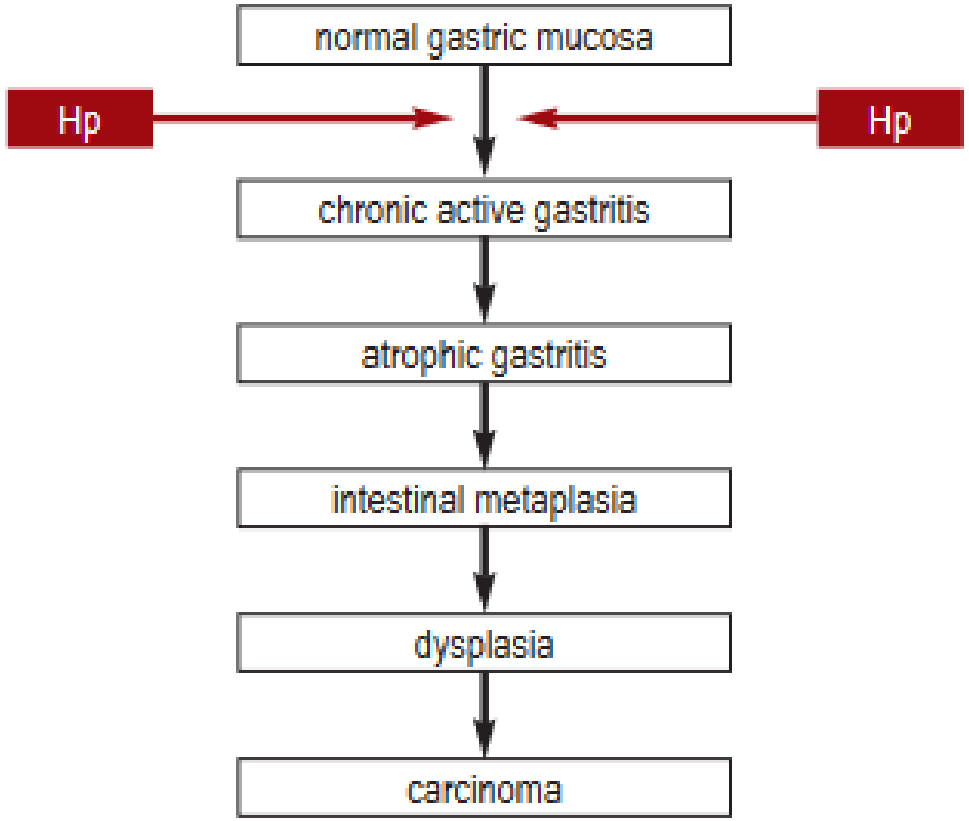
- Acquired during childhood
- Causes asymptomatic chronic infection
- Some H. pylori strains express specific genes conferring pro-inflammatory, cytotoxic and vacuolating properties
- Express Cytotoxin-associated antigen A (CagA) and Vacuolating cytotoxin A (VacA)
  - CagA - directly injected into the cytoplasm of epithelial cells, affecting cell morphology, proliferation and apoptosis
  - VacA - disrupts cell polarity, promotes epithelial cells apoptosis and inhibits T cell proliferation and effector function

GERD

- Gastroesophageal Reflux Disease

Gastric Dysfunction

- Genomic instability via reactive O and N species
- Induction of the immune system w/o infxn clearance
- Gastric and duodenal ulcers
- Mucosa associated tissue lymphoma
- Gastric adenocarcinoma



Gastric and Duodenal Ulcers

- Small portion of H. pylori infections
- Usually developed during late adulthood
- Approximately 1 in 5 peptic ulcers
  - Remainder related to NSAIDs
  - Eradicating H. pylori in NSAID users reduces the likelihood of peptic ulcers by one-half
- Urea breath test or stool antigen test is preferred
- First-line treatment – quadruple therapy plus PPI

Am Fam Physician. 2023 Feb;107(2):165-172.

Risks of Long Term PPI Use

- Fractures
- Interaction with antiplatelet medications
- Chronic kidney disease
- Clostridioides difficile infection
- Dementia
- Magnesium, calcium, and vitamin B12 micronutrient deficiencies

Systematic Review and Meta-Analysis

Halitosis and *helicobacter pylori* infection

A meta-analysis

Wenhuan Dou, MM, Juan Li, MM, Liming Xu, MM, Jianhong Zhu, PhD, Kewei Hu, MM, Zhenyu Sui, Jianzong Wang, MM, Lingling Xu, MM, Shaofeng Wang, MM , Guojian Yin, PhD

Abstract

Background: Halitosis is used to describe any disagreeable odor of expired air regardless of its origin. Numero have investigated the relation between *Helicobacter pylori* (*H pylori*) infection and halitosis, and even some re eradication have been prescribed to those patients with halitosis in the clinic. We conducted a meta-analysis to de between *H pylori* infection and halitosis.

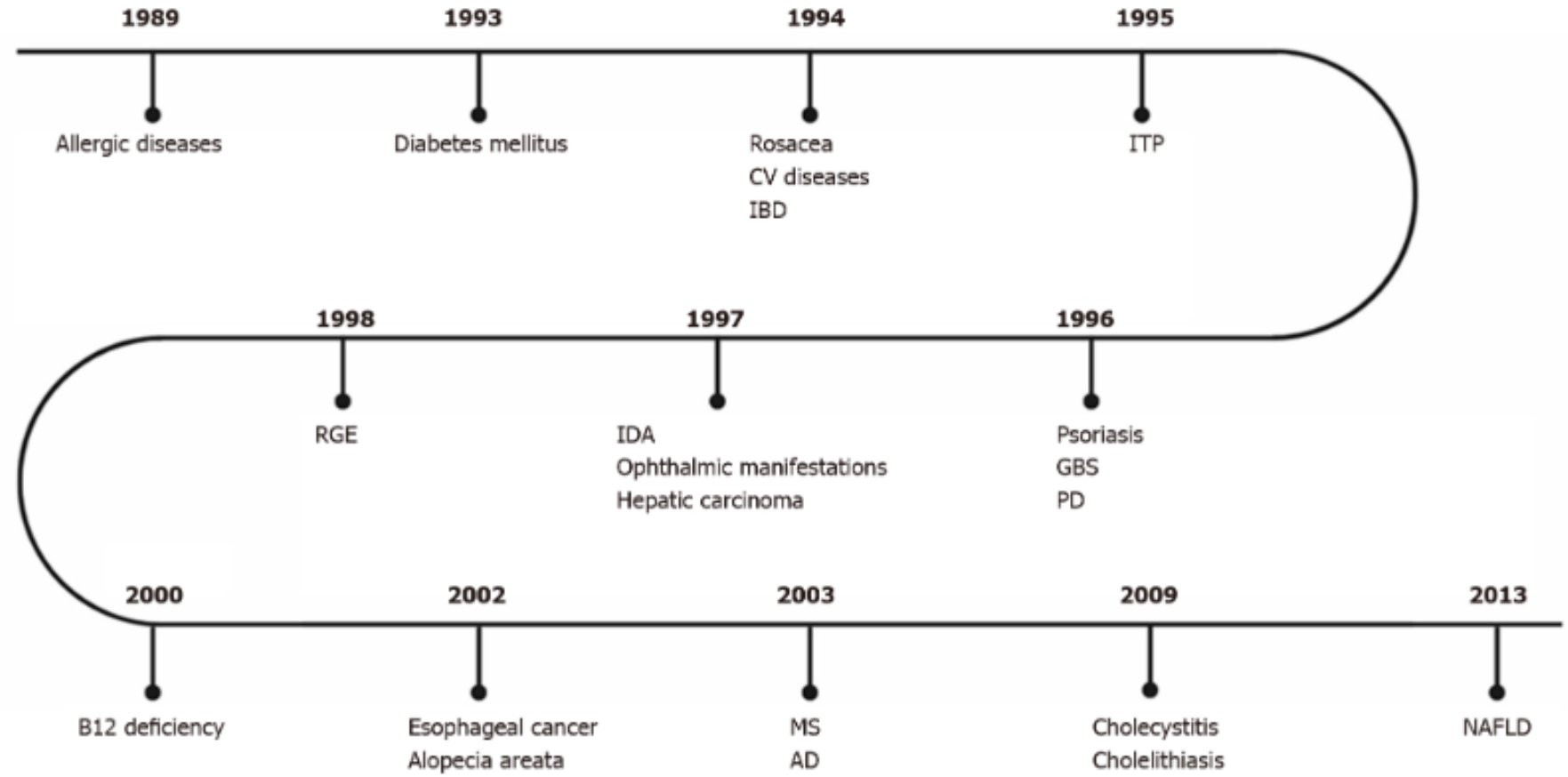
Objectives: To evaluate whether there is a real correlation between *H pylori* infection and halitosis, and whether *H* therapy will help relieve halitosis.

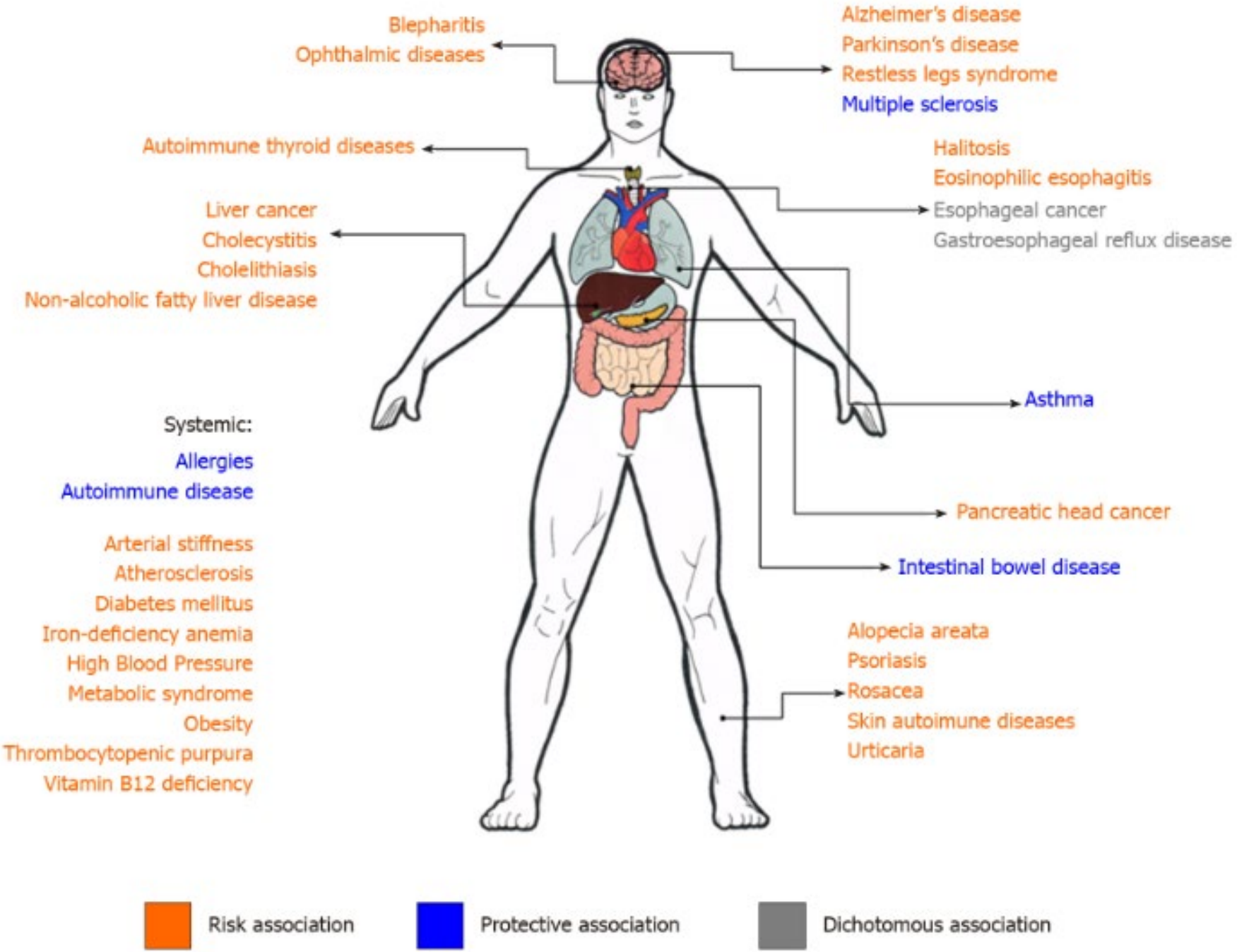
Methods: We searched several electronic databases (The Cochrane Library, MEDLINE, EMBASE, PubMed, We Wanfangdata) up to December 2015. Studies published in English and Chinese were considered in this review. studies was identified, the list of references reported in the included reports was reviewed to identify additional stu titles and abstracts, data extraction and quality assessment was undertaken independently and in duplicate. All a using Review Manager 5.2 software.

Results: A total of 115 articles were identified, 21 of which met the inclusion criteria and presented data that co analysis. The results showed that the OR of *H pylori* infection in the stomach between halitosis-positive patients and hali patients was 4.03 (95% CI: 1.41–11.50; $P=0.009$). The OR of halitosis between *H pylori*-positive patients and *H p* patients was 2.85 (95% CI: 1.40–5.83; $P=0.004$); The RR of halitosis after successful *H pylori* eradication in those *H p* halitosis-positive patients was 0.17 (95% CI: 0.08–0.39; $P<0.0001$), compared with those patients without succe eradication. And the RR of halitosis before successful *H pylori* eradication therapy was 4.78 (95% CI: 1.45–15. compared with after successful *H pylori* eradication therapy.

There is clear evidence that H pylori infection correlates with halitosis. H pylori infection might be important in the pathophysiological mechanism of halitosis, and H pylori eradication therapy may be helpful in those patients with refractory halitosis.

Extragastric Manifestations




Blepharitis
Ophthalmic diseases
Alzheimer's disease
Parkinson's disease
Restless legs syndrome
Multiple sclerosis
Autoimmune thyroid diseases
Halitosis
Eosinophilic esophagitis
Esophageal cancer
Gastroesophageal reflux disease
Liver cancer
Cholecystitis
Cholelithiasis
Non-alcoholic fatty liver disease
Asthma
Systemic:
Allergies
Autoimmune disease
Pancreatic head cancer
Intestinal bowel disease
Arterial stiffness
Atherosclerosis
Diabetes mellitus
Iron-deficiency anemia
High Blood Pressure
Metabolic syndrome
Obesity
Thrombocytopenic purpura
Vitamin B12 deficiency
Alopecia areata
Psoriasis
Rosacea
Skin autoimune diseases
Urticaria
Risk association
Protective association
Dichotomous association

Recalcitrant Nutrient Deficiencies

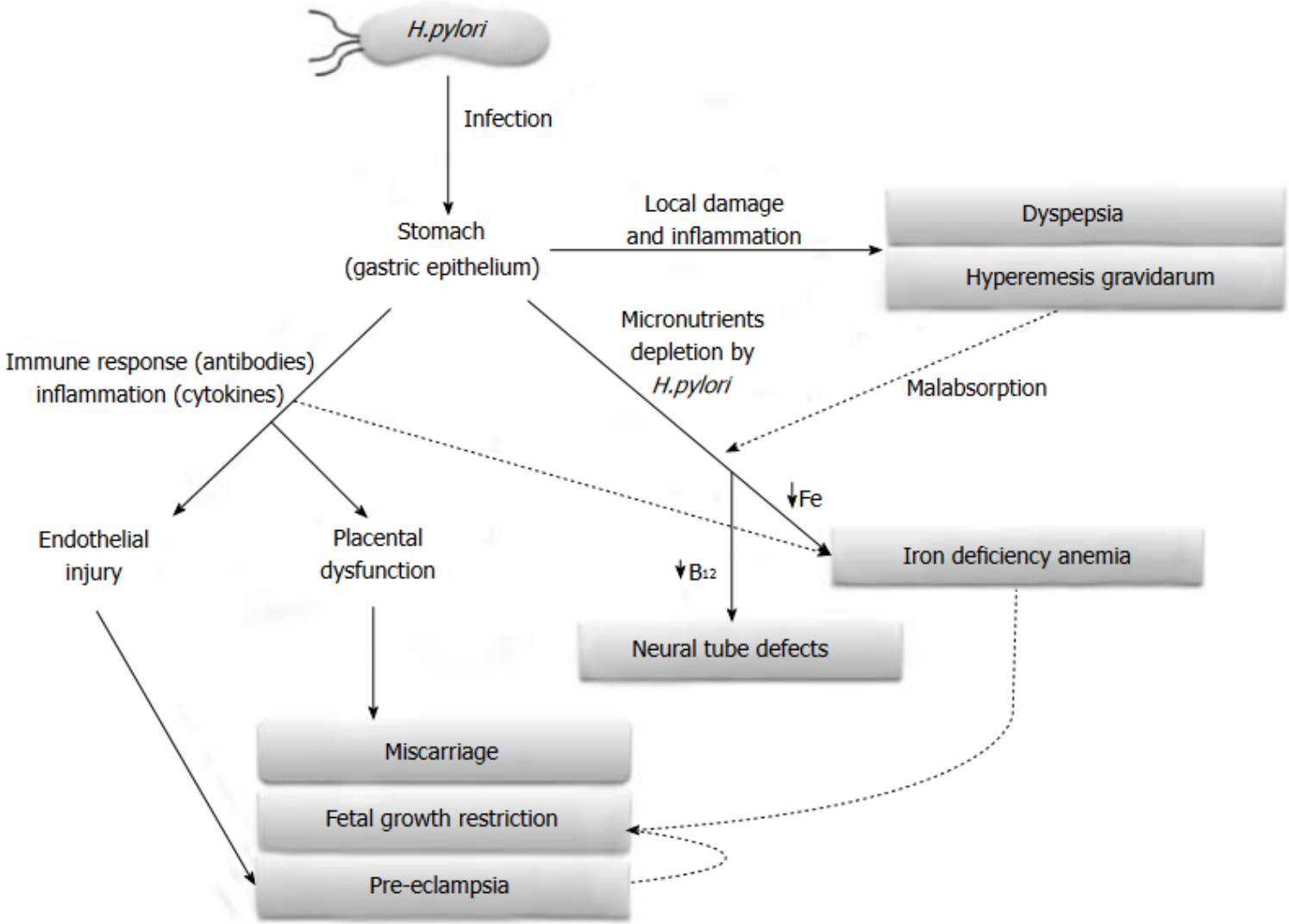
B12

- Pernicious anemia
- Antibiotic therapy shown to bring back serum B12 levels
- Elevations of homocysteine
  - Cardiovascular disease, neurodegeneration

Iron

- Anemia
- Linked to growth disorders in growth

Pregnancy



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TOPIC HIGHLIGHT

WJG 20th Anniversary Special Issues (6): *Helicobacter pylori*

Helicobacter pylori and pregnancy-related disorders

Simona Cardaropoli, Alessandro Rolfo, Tullia Todros

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Abstract

Helicobacter pylori (*H. pylori*) infection is investigated in gastric diseases even during pregnancy. In particular, this Gram-negative bacterium seems to be associated with hyperemesis gravidarum, a severe form of nausea and vomiting during pregnancy. During the last decade, the relationship among *H. pylori* and several extra-gastric diseases strongly emerged in literature. The correlation among *H. pylori* infection and pregnancy-related disorders was mainly focused on iron deficiency anemia, thrombocytopenia, fetal malformations, miscarriage, pre-eclampsia and fetal growth restriction. *H. pylori* infection may have a role in the pathogenesis of various pregnancy-related disorders through different mechanisms: depletion of micronutrients (iron and vitamin B_{12}) in maternal anemia and fetal neural tube defects; local or systemic induction of pro-inflammatory cytokines release and oxidative stress in gastrointestinal disorders and pre-eclampsia; cross-reaction between specific anti-*H. pylori* antibodies and antigens localized in placental tissue and endothelial cells (pre-eclampsia, fetal growth restriction, miscarriage). Since *H. pylori* infection is most likely acquired before pregnancy, it is widely believed that hormonal and immunological changes occurring during pregnancy could activate latent *H. pylori* with a negative impact not only on maternal health (nutritional deficiency, organ injury, death), but also on the fetus (insufficient growth, malformation, death) and sometime consequences can be observed later in life. Another important issue addressed by investigators was to determine whether it is possible to transmit *H. pylori* infection from mother to child and whether maternal anti-*H. pylori* antibodies could prevent infant's infection. Studies on novel diagnostic and therapeutic methods for *H. pylori* are no less important, since these are particularly sensitive topics in pregnancy conditions. It could be interesting to study the possible correlation between *H. pylori* infection and other pregnancy-related diseases of unknown etiology, such as gestational diabetes mellitus, obstetric cholestasis and spontaneous preterm delivery. Since *H. pylori* infection is treatable, the demonstration of its causative role in pregnancy-related disorders will have important social-economic implications.

Key words: *Helicobacter pylori*; Pregnancy; Hyperemesis gravidarum; Iron deficiency anemia; Pre-eclampsia; Fetal growth restriction; Gastrointestinal disorders

Core tip: *Helicobacter pylori* (*H. pylori*) infection in pregnancy is not only associated with gastrointestinal disorders such as hyperemesis gravidarum, but also with iron deficiency anemia, fetal malformations, miscarriage, pre-eclampsia and fetal growth restriction. These pregnancy related-disorders are potentially life-threatening for both mother and fetus/neonate. Another important issue that has been addressed in literature was the question of whether it is possible to transmit *H. pylori* infection from mother to child and whether maternal anti-*H. pylori* antibodies could prevent infant's infection. Indeed, if *H. pylori* is actually a causal factor, the public health implications would be important since the infection is treatable.

Cardaropoli S, Rolfo A, Todros T. *Helicobacter pylori* and pregnancy-related disorders. *World J Gastroenterol* 2014; 20(3): 654-664

Halting the Cycle of Small Intestinal Bacterial Overgrowth

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Review > Gastroenterol Clin North Am. 2020 Sep;49(3):571-587. doi: 10.1016/j.gtc.2020.04.010. Epub 2020 Jun 14.

Small Intestinal Bacterial Overgrowth: How to Diagnose and Treat (and Then Treat Again)

Brian Ginnebaugh [1], William D Chey [2], Richard Saad [2]

Review > Crit Rev Oncog. 2020;25(4):365-379. doi: 10.1615/CritRevOncog.2020036017.

Small Intestinal Bacterial Overgrowth: A Critical Review of an Underrecognized but Disrupting Entity

Small intestinal bacterial overgrowth (SIBO) is a common gastrointestinal (GI) problem, but its diagnosis is often missed in the clinical setting.

Symptomatic relief can be achieved through multiple antibiotics regimens, but correction of underlying etiology, if possible, is necessary for long-lasting cure.

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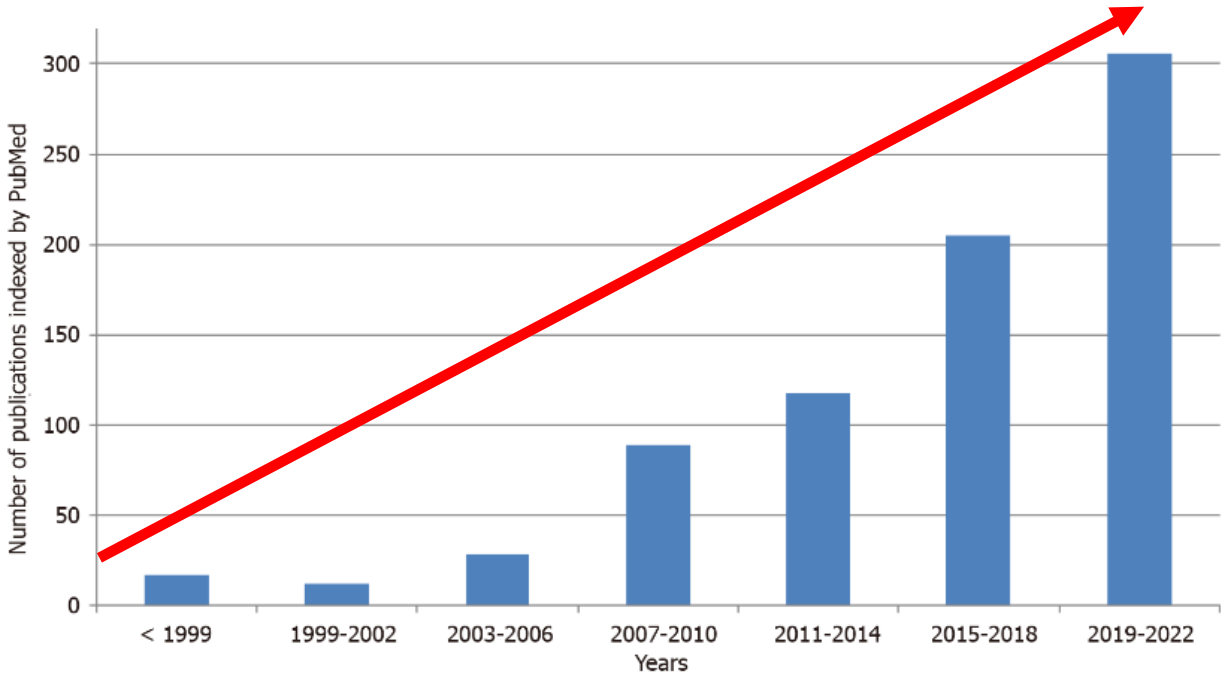

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Figure 1 Number of publications indexed by PubMed on small intestinal bacterial overgrowth by year.

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REVIEW

Epidemiology of small intestinal bacterial overgrowth

Irina Efremova, Roman Maslennikov, Elena Poluektova, Ekaterina Vasilieva, Yury Zharikov, Andrey Suslov, Yana Letyagina, Evgenii Kozlov, Anna Levshina, Vladimir Ivashkin

Specialty type: Gastroenterology and hepatology

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Recurrent SIBO

- Approximately 44% of patients with SIBO may experience a relapse of symptoms within 9 months of initial treatment
- Approximately 30%–40% of patients may not have resolution of SIBO symptoms with antibiotic trials
  - Norfloxacin, Broad-spectrum antibiotics, minocycline, Ciprofloxacin, metronidazole, neomycin, amoxicillin

Clinical and Translational Gastroenterology 2019;10:e00078.
Am J Gastroenterol 2008; 103:2031–5.

SIBO Roadmap

- Clinical features
- Testing
- Health implications
- Conventional guidelines
- Complementary approaches
- Probiotics?

SIBO Defined

- Presence of an abnormal number of bacteria in the small intestine
- Greater than 10^5 CFU/mL (Less than 10^3 CFU/mL is normal)
- Colonic-type bacteria
- Shift from Gram-positive to Gram-negative dominant anaerobes (aerobes still present)
- Constellation of GI symptoms (often unexplained)
- Symptoms consistent with irritable bowel syndrome

Small Intestinal Bacterial Overgrowth: Clinical Features and Therapeutic Management

Satish S. C. Rao, MD, PhD[1] and Jigar Bhagatwala, MBBS, MPH[1]

Small intestinal bacterial overgrowth (SIBO) is a common, yet underrecognized, problem. Its prevalence is unknown because SIBO requires diagnostic testing. Although abdominal bloating, gas, distension, and diarrhea are common symptoms, they do not predict positive diagnosis. Predisposing factors include proton-pump inhibitors, opioids, gastric bypass, colectomy, and dysmotility. Small bowel aspirate/culture with growth of 10^3–10^5 cfu/mL is generally accepted as the "best diagnostic method," but it is invasive. Glucose or lactulose breath testing is noninvasive but an indirect method that requires further standardization and validation for SIBO. Treatment, usually with antibiotics, aims to provide symptom relief through eradication of bacteria in the small intestine. Limited numbers of controlled studies have shown systemic antibiotics (norfloxacin and metronidazole) to be efficacious. However, 15 studies have shown rifaximin, a nonsystemic antibiotic, to be effective against SIBO and well tolerated. Through improved awareness and scientific rigor, the SIBO landscape is poised for transformation.

Clinical and Translational Gastroenterology 2019;10:e00078. https://doi.org/10.14309/ctg.0000000000000078

INTRODUCTION

The adult gastrointestinal (GI) tract has the largest microbial population in the human body (1); the predominant site is the colon, containing 38 trillion bacteria (2). Culture-independent methods, such as next-generation sequencing, show low concentration of distinct bacterial populations in the duodenum of healthy individuals, in contrast with bacterial populations that inhabit the mouth (3). Bacterial concentrations increase progressively along the small intestine (4,5).

Small intestinal bacterial overgrowth (SIBO) is characterized by the presence of an abnormal amount of bacteria in the small intestine together with a constellation of GI symptoms. The purpose of this article is to provide an up-to-date review of SIBO, including symptom patterns, predisposing risk factors, prevalence, specialized diagnostic testing, and potential therapeutic interventions, and to describe gaps in our knowledge and unmet needs.

METHODS

A PubMed search was performed on June 8, 2018, to identify English-language publications of clinical trials pertaining to SIBO in adults since 1985 using the search terms "small bowel bacterial overgrowth," "small intestinal bacterial overgrowth," "SIBO," "epidemiology," "diagnosis," "treatment," "antibiotic (e.g., ciprofloxacin, cotrimoxazole, and metronidazole)," "rifaximin," or "probiotic." Clinical studies of rifaximin ($n = 15$), systemic antibiotics ($n = 6$), and probiotics ($n = 3$) in SIBO were included, whereas studies of combination therapies, for example, rifaximin with another antibiotic and/or other combination of systemic antibiotics or probiotics, were excluded from this review. A total of 23 references on predisposing factors and 4 on diagnostic testing for SIBO were included. Although we recognize that SIBO occurs in a wide spectrum of diseases discussed below, most literature on this topic has focused on patients presenting with either unexplained symptoms or symptoms of irritable bowel syndrome (IBS). Our review primarily focuses on these patients, as they are most commonly encountered in gastroenterology clinics, but other conditions are appropriately referenced wherever necessary.

CLINICAL FEATURES, PREVALENCE, AND PATHOETIOLOGY

Symptoms of SIBO are nonspecific and include abdominal pain, belching, bloating, diarrhea, distension, flatulence, and indigestion that overlap and vary in frequency, duration, and severity. Typically, over two-thirds of patients report the aforementioned symptoms (6,7). Diagnosis of SIBO is challenging, as illustrated by 1 study in which mean total symptom scores were similar regardless of whether patients tested positive or negative with duodenal aspirate and breath testing ($P = 0.9$) (6). Because a SIBO diagnosis requires specialized testing (e.g., microbial culture and breath testing), and owing to variability in patient populations and methods used to establish a diagnosis across studies (8), prevalence has been difficult to estimate. However, SIBO appears to be more prevalent in women and in older individuals (9).

Several factors are associated with or predispose patients to SIBO, including small intestinal dysmotility (10). A study using duodenal aspirate/culture demonstrated that patients with small intestinal dysmotility were at increased risk of SIBO (>10^3 colony-forming units [cfu]/mL threshold, odds ratio [OR], 3.6; $P = 0.0003$; >10^5 cfu/mL threshold, OR, 2.7; $P = 0.005$) (7).

Predisposing Factors

Table 1. Predisposing factors for SIBO

Category	Factor
Demographics	Female sex (9)
	Age (9)
Medication use	PPIs (7,18,19)
	Opioids (23)
GI conditions	Dyspepsia (24)
	IBD (9,25)
	IBS (11–14)
	Intestinal dysmotility (7)
	Small bowel diverticula (9)
	Systemic sclerosis (33)
Other conditions	Abdominal surgery (i.e., hysterectomy, gastrectomy, cholecystectomy, colectomy, and Roux-en-Y gastric bypass) (17,26,34)
	Coronary artery disease (16)
	Diabetes (27)
	Hypothyroidism (28)
	Pancreatitis (9,29)
	Parkinson's disease (30)
	Restless legs syndrome (31)
	Rosacea (32)

GI, gastrointestinal; IBD, inflammatory bowel disease; IBS, irritable bowel syndrome; PPI, proton-pump inhibitor; SIBO, small intestinal bacterial overgrowth.

Diagnosis

Symptoms

- Bloating
- Gas
- Distention
- Abdominal pain
- Diarrhea

Non-Invasive

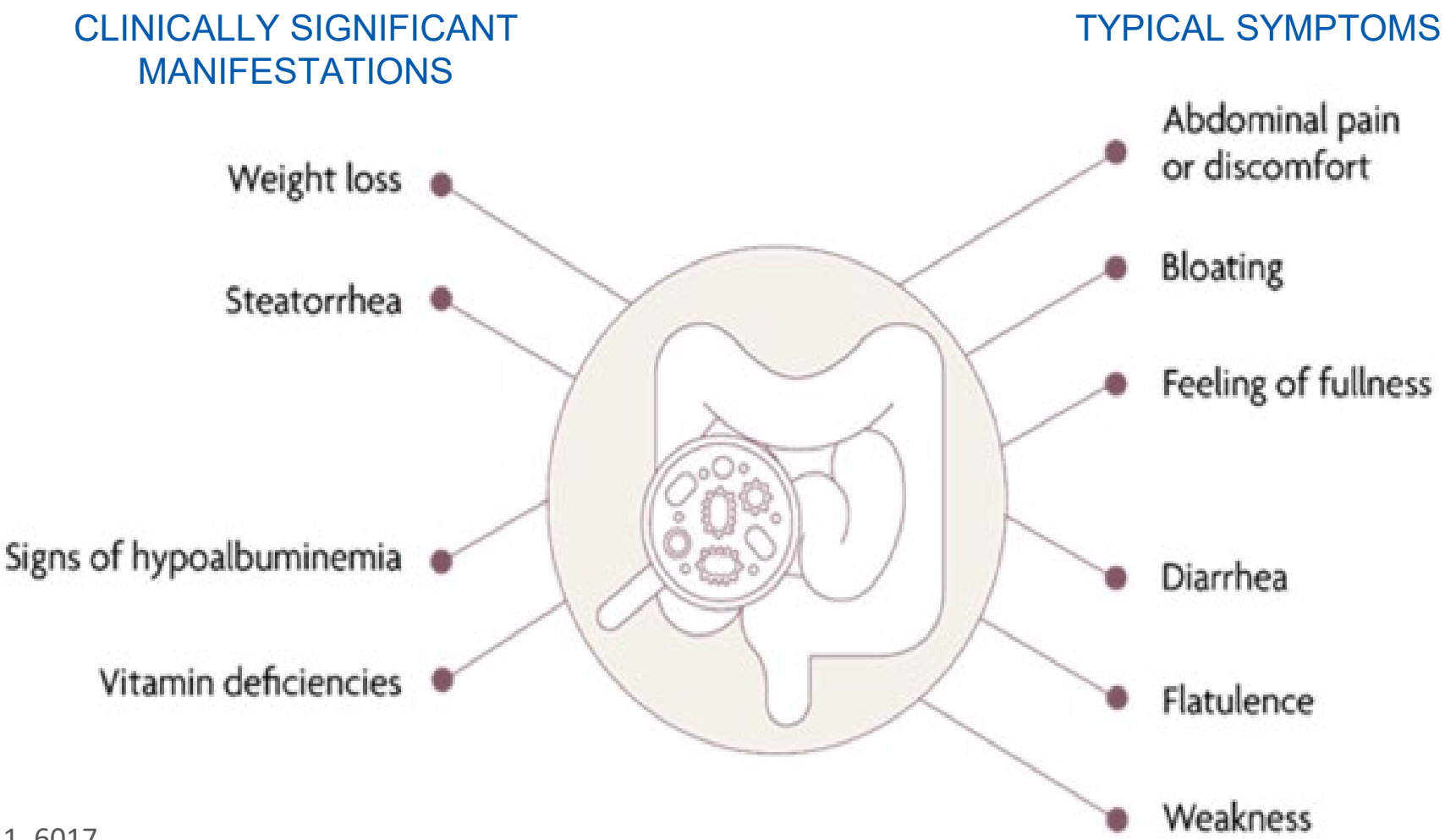
- Glucose Breath Test
- Lactulose Breath Test

Invasive

- Small Bowel Aspirate

Diagnostics

Clinical Picture



J. Clin. Med. 2022, 11, 6017.

Table 1. Symptoms implicated with small intestinal bacterial overgrowth and possible mechanisms (116,117)

Symptom	Possible mechanisms
Bloating	Multifactorial: increased gas production from bacterial fermentation, visceral hypersensitivity, and decreased small intestinal elasticity and/or transit
Diarrhea	Multifactorial: bacterial digestion produces gas and osmotically active byproducts, bacteria and byproducts injure mucosa resulting in increased water output, resulting in lactase deficiencies, and bacterial deconjugation of bile salts interferes with fat absorption
Abdominal pain	Multifactorial: brain-gut, visceral hypersensitivity, and decreased small intestinal elasticity and/or transit
Constipation	Methane production slows intestinal transit; increased gas distension interferes with intestinal contractions
Anemia/neuropathy	Bacterial consumption of vitamin B12
Inability to gain weight and weight loss	Reduced availability of nutrients due to bacterial digestion
Steatorrhea/fat-soluble vitamin deficiencies	Bacterial deconjugation of bile acids resulting in insufficient absorption of fats and fat-soluble vitamins
Systemic reactions (i.e., brain fog, anxiety/depression, and dermatologic conditions)	Multifactorial: increased bacterial counts and intestinal barrier destruction can result in hypersensitivity reactions/immune response; vitamin deficiencies; bacteria and byproducts may traverse the disrupted intestinal barrier
Distension	Multifactorial: increased gas production from bacterial fermentation; decreased small intestinal elasticity and/or transit

Clinical and Translational Gastroenterology 2023;14:e00567.

Assessment Options

- Direct culture of small intestinal contents – Gold Standard for SIBO
  - Cumbersome, invasive, costly, and subject to some contention regarding diagnostic thresholds of bacterial counts, variable sampling, inconsistent processing techniques
- Breath test – Hydrogen, Methane
  - Noninvasive, inexpensive, and relatively straightforward
  - Lack sensitivity and specificity
- Stool testing – Depends on the methodology
- Empiric therapeutic trial of antibiotics / antimicrobials

Breath Test Substrates

- Glucose – absorbed in the proximal small intestine
- Lactulose – unabsorbed in the small intestine, passes into the colon
- Differing spatial and temporal reactions with the gut microbiota
- Lactose and Fructose - Substrates for detection of malabsorption, not SIBO

Hydrogen-Methane Test Execution

- Appropriate test preparation required
- Diet – low-residue, bland, limited fat, 12 hour fast
- Medications – duration of effects of PPIs, opioids, antibiotics
    - Variable washout period
- Lifestyle – smoking, alterations of gases
- Substrates – inconsistent load administration, lack of agreement on substrate choice
    - Glucose generally preferred
- Inability to obtain a baseline standard due to patient physiological factors
    - Oral flora, oral antiseptic use

Test Preparation

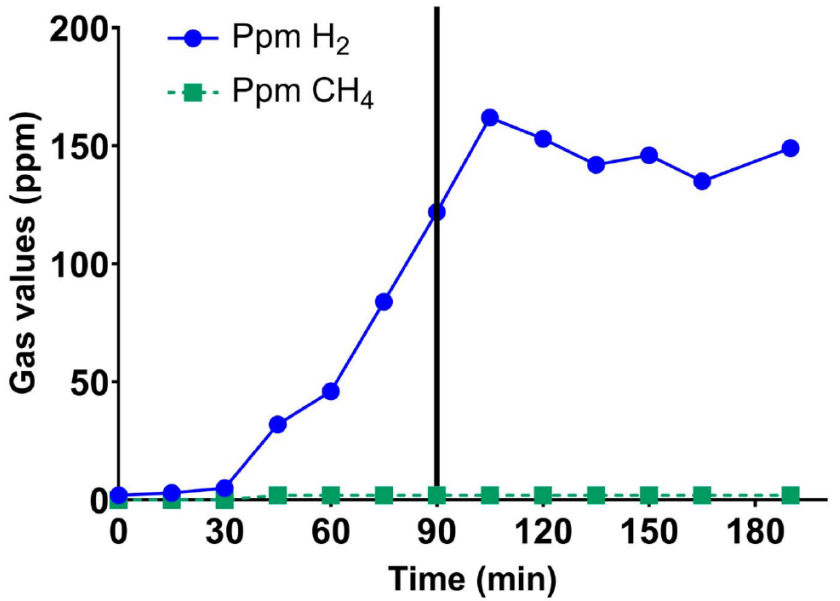
Table 1. Recommended preparation for breath testing.

Period before the Breath Test	Drugs/Activities to Be Avoided
4 weeks	Oral or intravenous antibiotics Prokinetic agents
2 weeks	Probiotics
1 week	Proton pump inhibitors
48 h	Motility regulators: loperamide, metoclopramide, trimebutine
24 h	Alcohol Fiber (particularly non-soluble fiber)
12 h	Oral food intake (only water is allowed)
The morning on the day of the test	Smoking Physical exertion Food Regularly used medications are allowed

J. Clin. Med. 2022, 11, 6017.

Breath Test Results

	Sample	Ppm H_2	Ppm CH_4	Concentration CO_2 (%)	(f) CO_2
Baseline	1	2	0	4.0	1.38
15 min	2	3	0	3.7	1.49
30 min	3	5	0	3.8	1.45
45 min	4	32	0	3.8	1.45
60 min	5	46	0	3.9	1.41
75 min	6	84	0	4.0	1.38
90 min	7	122	0	4.3	1.28
105 min	8	162	0	3.9	1.41
120 min	9	153	0	4.1	1.34
135 min	10	142	0	4.1	1.34
150 min	11	146	0	4.1	1.34
165 min	12	135	0	3.9	1.41
190 min	13	149	0	4.2	1.31



Hydrogen - >20ppm after 90 mins
Methane - >10ppm anytime

Sensitivity and Specificity

Table 3. Test characteristics of glucose and lactulose breath tests based on published data using jejunal aspirates as the gold standard (54)

Test characteristics	Glucose breath test (668 patients, 14 studies)	Lactulose breath test (214 patients, 4 studies)
Pooled sensitivity (True Positive with Condition)	54% (48%–61%)	42% (32%–53%)
Pooled specificity (True Negative without Condition)	83% (79%–87%)	71% (62%–78%)
Pooled positive likelihood ratio	2.45 (1.51–3.97)	1.30 (0.77–2.22)
Pooled negative likelihood ratio	0.60 (0.45–0.80)	0.79 (0.57–1.08)
Pooled diagnostic odds ratio	5.17 (2.42–11.05)	1.77 (0.72–4.37)
Area under the curve of summary receiver operating characteristic (SROC) curve	0.7418	0.5582

Test with Understanding

We found no significant correlation of glucose breath test results with either the number of bacterial colonies or with the DNA-based bacterial cell counts.

Neurogastroenterol Motil. 2018 Nov;30(11):e13350.

Assessment Anatomy

- The small bowel is 19 ft long, and can be colonized anywhere
- Glucose when administered orally is absorbed within the first 3–6 ft of small bowel
- Duodenal aspirates are typically from the proximal 2 ft of small bowel
- Relying on glucose breath testing alone to make a diagnosis of SIBO will provide a low yield of about 33%
- Negative GBT is not confirmation of absence of SIBO

Clinical and Translational Gastroenterology (2018) 9:194

Review > Curr Opin Gastroenterol. 2023 May 1;39(3):211-218.
doi: 10.1097/MOG.0000000000000928. Epub 2023 Mar 1.

Unravelling the controversy with small intestinal bacterial overgrowth

Summary: As a first step to precisely characterize the potential link between SIBO and various disorders, we need to address the methodological limitations of the available traditional tests for diagnosing SIBO.

Additional Lab Findings

- Megaloblastic anemia
- Iron-deficiency anemia
- Fat-soluble vitamin (A, D, and E) deficiencies
- Vitamin B12 deficiency
- Hypoalbuminemia

- Elevated CRP
- Elevated AST
- Hyperlipidemia
- Nutrient deficiencies

SIBO Bacterial Species

- *Streptococcus*
- *Staphylococcus*
- *Bacteroides*
- *Lactobacillus*
- *Enterobacteriaceae* family
  - *Escherichia*
  - *Klebsiella*
  - *Proteus* genera

COMMENSAL BACTERIA	Result	Reference
Bacteroides fragilis	1.31e11	1.6e9 - 2.5e11
Bifidobacterium spp.	1.09e10	> 6.7e7
Enterococcus spp.	2.71e7	1.9e5 - 2.0e8
Escherichia spp.	1.45e9	3.7e6 - 3.8e9
Lactobacillus spp.	8.82e7	8.6e5 - 6.2e8
Enterobacter spp.	4.43e6	1.0e6 - 5.0e7
Akkermansia muciniphila	8.16e5	1.0e1 - 8.2e6
Faecalibacterium prausnitzii	1.35e6	1.0e3 - 5.0e8
Roseburia spp.	6.65e8	5.0e7 - 2.0e10

DYSBIOTIC & OVERGROWTH BACTERIA	Result		Reference
Bacillus spp.	1.25e6		< 1.76e6
Enterococcus faecalis	2.98e3		< 1.00e4
Enterococcus faecium	7.42e3		< 1.00e4
Morganella spp.	<dl		< 1.00e3
Pseudomonas spp.	<dl		< 1.00e4
Pseudomonas aeruginosa	<dl		< 5.00e2
Staphylococcus spp.	<dl		< 1.00e4
Staphylococcus aureus	1.02e3	High ↑	< 5.00e2
Streptococcus spp.	1.22e4	High ↑	< 1.00e3

Microorganisms 2023, 11, 573.

Two Subtypes
Well-Established

- Upper aerodigestive tract SIBO
    - Caused predominantly by oral cavity bacteria, including *Prevotella sp.* and *Streptococcus viridans*
- Coliform SIBO
    - Characterized predominantly by bacteria from the distal segments of the gastrointestinal tract, such as *Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis, Enterococcus sp.*, or *Clostridioides sp.*
- Despite the distinction between the two types, their importance in clinical practice is limited due to the similar symptoms and treatment.

J. Clin. Med. 2022, 11, 6017.
J. Clin. Gastroenterol. 2020, 54, 150–157
Clin. Transl. Gastroenterol. 2019, 10, e00073

When it Matters

Clinical Presentation of Small Intestinal Bacterial Overgrowth from Aerodigestive Tract Bacteria Versus Colonic-Type Bacteria: A Comparison Study

Daanish A. Siddique, Claire L. Jansson-Knodell, Anita Gupta, Gage Howard, Matthew E. Bohm, Robert M. Siwiec, David E. Nelson, Andrea S. Shin & John M. Wo

Digestive Diseases and Sciences **68**, 3390–3399 (2023) | Cite this article

We found differences in **iron deficiency** and **underlying risk factors** between ADT and colonic-type SIBO. However, distinct clinical profiles remained elusive. Future research is needed to develop validated symptom assessment tools and distinguish cause from correlation.

An Additional Word About Methane

- Methane impacts GI function
- Methane more directly related to constipation
- More recent acknowledgement of Intestinal Methanogen Overgrowth
- Methanogens are actually Archaea, not bacteria
- Methanogens are found in the small intestine and colon
- Do not respond as well to single antibiotics

Clinical and Translational Gastroenterology 2023;14:e00567.

Which One?

1. Small Intestinal Bacterial Overgrowth
2. Large Intestinal Bacterial Overgrowth
3. Small Intestinal Fungal Overgrowth
4. Intestinal Methanogen Overgrowth

microorganisms

MDPI

Review

Association between Gut Dysbiosis and the Occurrence of SIBO, LIBO, SIFO and IMO

Michalina Banaszak [1,2], Ilona Górna [1], Dagmara Woźniak [1,2], Juliusz Przysławski [1] and Sławomira Drzymała-Czyż [1,*]

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Abstract: Gut microbiota is the aggregate of all microorganisms in the human digestive system. There are 10^{14} CFU/mL of such microorganisms in the human body, including bacteria, viruses, fungi, archaea and protozoa. The Firmicutes and Bacteroidetes bacteria phyla comprise 90% of the human gut microbiota. The microbiota support the healthy functioning of the human body by helping with digestion (mainly via short-chain fatty acids and amino acids) and producing short-chain fatty acids. In addition, it exhibits many physiological functions, such as forming the intestinal epithelium, intestinal integrity maintenance, the production of vitamins, and protection against pathogens. An altered composition or the number of microorganisms, known as dysbiosis, disrupts the body's homeostasis and can lead to the development of inflammatory bowel disease, irritable bowel syndrome, and metabolic diseases such as diabetes, obesity and allergies. Several types of disruptions to the gut microbiota have been identified: SIBO (Small Intestinal Bacterial Overgrowth), LIBO (Large Intestinal Bacterial Overgrowth), SIFO (Small Intestinal Fungal Overgrowth), and IMO (Intestinal Methanogen Overgrowth). General gastrointestinal problems such as abdominal pain, bloating, gas, diarrhoea and constipation are the main symptoms of dysbiosis. They lead to malabsorption, nutrient deficiencies, anaemia and hypoproteinaemia. Increased lipopolysaccharide (LPS) permeability, stimulating the inflammatory response and resulting in chronic inflammation, has been identified as the leading cause of microbial overgrowth in the gut. The subject literature is extensive but of limited quality. Despite the recent interest in the gut microbiome and its disorders, more clinical research is needed to determine the pathophysiology, effective treatments, and prevention of small and large intestinal microbiota overgrowth. This review was designed to provide an overview of the available literature on intestinal microbial dysbiosis (SIBO, LIBO, SIFO and IMO) and to determine whether it represents a real threat to human health.

Citation: Banaszak, M.; Górna, I.; Woźniak, D.; Przysławski, J.; Drzymała-Czyż, S. Association between Gut Dysbiosis and the Occurrence of SIBO, LIBO, SIFO and IMO. *Microorganisms* 2023, *11*, 573. https://doi.org/10.3390/

SIFO - Fungus

- Typically, the result of a weakened immune system
    - HIV, antibiotics, cancer, chemotherapy, diabetes, immunosuppressants, steroids
- Similar symptoms to SIBO
- Can be part of a larger systemic fungal infection
- *Candida albicans* is the most common organism
- Limited benefit of testing, requires minimal threshold to be antibody positive
- Treatment – lacks establishment

Intestinal Methanogen Overgrowth

- Most recent addition to dysbiosis characterization
- Not bacteria, but rather Archaea
- Archaea – methane producing organisms, use hydrogen
- Overgrowth possible in the colon and throughout the entire body
- Less likely to exhibit B12 deficiency
- Diagnosis – breath test, >10ppm
- Treatment with antibiotics is recommended

Small Intestinal Bacterial Overgrowths and Intestinal Methanogen Overgrowths Breath Testing in a Real-Life French Cohort

Anne Plauzolles, PhD[1], Stella Uras, Master degree[1,2], Guillaume Pénaranda, PhD[1], Marion Bonnet, PharmD, PhD[1], Patrick Dukan, MD[3], Frédérique Retornaz, MD[3,*] and Philippe Halfon, MD, PharmD, PhD[1,3,*]

INTRODUCTION: **Breath testing has become a widely used tool to diagnose small intestinal bacterial overgrowths (SIBOs) and intestinal methanogen overgrowths (IMOs) in clinical settings. Owing to the heterogeneity in clinical manifestations and lack of standardization among centers performing breath testing, SIBO and IMO can be easily overlooked by the clinician. We studied the prevalence and symptoms of SIBO/IMO in French patients referred for breath testing after seeking medical advice.**

There are little discriminating symptoms that can help the clinician to identify patients likely to have a SIBO/IMO.

with a positive test except for joint pain that was less prevalent among LBT positive patients ($P = 0.038$). In 86.5% of IMOs, positivity with CH_4 values ≥10 ppm could be identified at baseline.

DISCUSSION: **There are little discriminating symptoms that can help the clinician to identify patients likely to have a**

COMMENSAL OVERGROWTH MICROBES		
Desulfovibrio spp.	2.96e7	< 7.98e8
Methanobacteriaceae (family) ←	1.07e8	< 3.38e8

SUPPLEMENTARY MATERIAL accompanies this paper at http://links.lww.com/CTG/A898

Clinical and Translational Gastroenterology 2023;14:e00556. https://doi.org/10.14309/ctg.0000000000000556

Extra-Intestinal Associations

- Irritable Bowel Syndrome
- Inflammatory Bowel Syndrome
- Celiac Disease
- Hepatic Dysfunction
- Heart Failure
- Diabetes
- Scleroderma
- Parkinson Disease
- Hypothyroidism
- Restless Leg Syndrome

Irritable Bowel Syndrome

- Commonly confused with SIBO
- Overlapping Symptoms
- SIBO is significantly more prevalent in IBS (C, D & A) compared to controls
  - IBS-D more common than IBS-C
- SIBO secondary to IBS interventions
  - Proton pump inhibitors
  - Narcotics – inhibiting gut motility
  - Multiple medications – increased likelihood with age, reduced motility

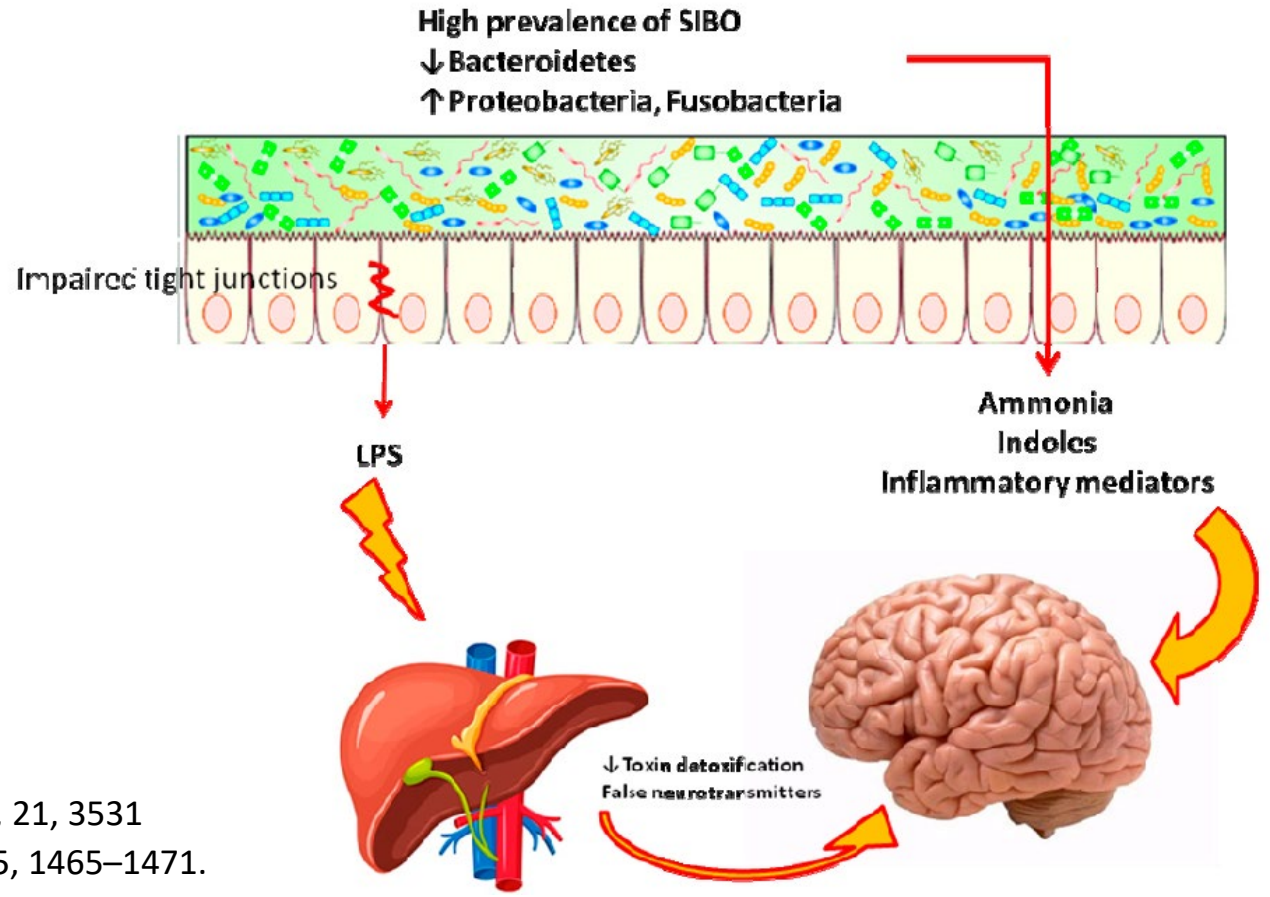
Inflammatory Bowel Disease
(Crohn's disease and Ulcerative Colitis)

- 9.51 times higher risk of SIBO in IBD than in controls
- Predisposing factors
  - Intestinal surgery (ICV resection), slower transit time
- May be a primary cause of flares
- Increased calprotection (local inflammatory marker)
  - Not as likely with IBS
- Antimicrobials may be more effective than immunosuppresants
- Consider when immune focused strategies are not as effective as anticipated

Int. J. Mol. Sci. 2020, 21, 3531
Aliment. Pharmacol. Ther. 2019, 49, 624–635.

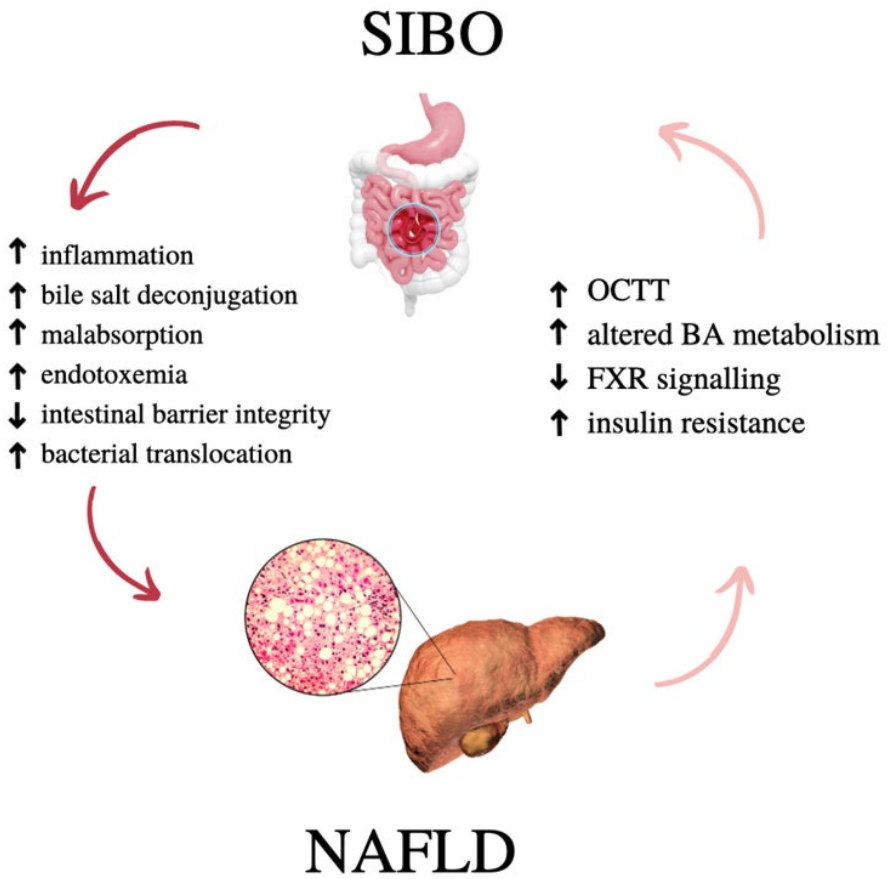
Hepatic Dysfunction

- Increased risk of SIBO in cirrhosis
  - SIBO as a consequence and potentiator of cirrhosis
  - Altered transit time
- Increased ammonia accumulation leading to hepatic encephalopathy
- Resultant endotoxemia
- Decreased Bacteroidetes
- NAFLD



Int. J. Mol. Sci. 2020, 21, 3531
Dig. Dis. Sci. 2010, 55, 1465–1471.

NAFLD
Non-Alcoholic Fatty Liver Disease



Nutrients 2023, 15, 1323.

World Journal of Gastroenterology

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World J Gastroenterol 2022 March 14; 28(10): 1067-1077

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ISSN 1007-9327 (print) ISSN 2219-2840 (online)

ORIGINAL ARTICLE

Observational Study

Gut dysbiosis and small intestinal bacterial overgrowth as independent forms of gut microbiota disorders in cirrhosis

Roman Maslennikov, Vladimir Ivashkin, Irina Efremova, Elena Poluektova, Anna Kudryavtseva, George Krasnov

Cirrhosis and SIBO

- Portal hypertension in liver cirrhosis substantially changes the intraluminal milieu of the gut.
- Liver cirrhosis is an independent risk factor for SIBO.
- Small intestinal motility disorder, especially slow transit in advanced liver disease may partake in SIBO.
- SIBO was diagnosed in 50%-60% of patients with liver cirrhosis.

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World J Gastroenterol. 2010 Jun 28; 16(24): 2978–2990.

Diabetes

- SIBO is more frequent in diabetic patients
- Contributing factors of pathogenesis
    - Visceral neuropathy
    - Slower transit time
    - Reduced exocrine pancreatic function
        - Decreased trypsin, decreased defensin, decreased pancreatic antibacterial activity

Rheumatological Diseases
(Scleroderma)

- Altered function of the enteric nervous system or visceral smooth muscle layer
- Onset of dysmotility and reduced small intestinal clearance
- Small intestinal functional impairment due to fibrosis and microvascular changes

Dermatological Diseases

- The human microbiota ecosystem may impact on cutaneous physiology and pathology
  - Indirectly - Regulating the immune system
  - Directly -Transfer of gut microbiome and their metabolites to the skin
- Rosacea
  - Onset may be triggered by SIBO by production of inflammatory cytokines
- Psoriasis
- Role of H. pylori as a precursor to SIBO
- Gut-skin axis disruption

Parkinson's Disease

- Common symptoms included impaired gut motility, constipation, delayed gastric emptying
- Inherently favors dysbiosis and SIBO
- Prevalence of up to 67%
- Positive SIBO associated with unpredictable fluctuations, rigidity and worse indexes of motor dysfunction
- Alters drug absorption (levodopa) and neuro-inflammation
- Eradication may lead to motor improvement

Subclinical Hypothyroidism

In addition, the TPOAb-positive rate and TSH levels were higher but the FT4 level was lower in SIBO-positive patients compared to SIBO-negative patients in study group.

Front Endocrinol (Lausanne). 2021 May 24;12:604070.

frontiers in Endocrinology

ORIGINAL RESEARCH
published: 24 May 2021
doi: 10.3389/fendo.2021.604070

Small Intestinal Bacterial Overgrowth in Subclinical Hypothyroidism of Pregnant Women

Biao Wang, Yajuan Xu, Xiaofeng Hou, Jingjing Li, Yanjun Cai, Yingqi Hao, Qian Ouyang, Bo Wu, Zongzong Sun, Miao Zhang and Yanjie Ban*

Department of Obstetrics and Gynecology, The Third Affiliated Hospital of Zhengzhou University, Zhengzhou, China

Objective: To evaluate the small intestinal bacterial overgrowth (SIBO) of subclinical hypothyroidism of pregnant women, and explore their possible relevance.

Methods: In total, 224 pregnant women with subclinical hypothyroidism during pregnancy (study group) and 196 pregnant women whose thyroid function was normal (control group) were enrolled in this study. Lactulose-based hydrogen and methane breath test was performed to evaluate the growth of intestinal bacteria. The serum-free thyroid hormone (FT4), thyroid-stimulating hormone (TSH), thyroid peroxidase antibody (TPOAb), body mass index (BMI) and gastrointestinal symptoms were detected and recorded.

Results: The positive rates of SIBO were 56.7% and 31.6% in study group and control group, respectively. The levels of C response protein (CRP), abdominal distension and constipation in study group were higher than those in the control group. The risk of abdominal distension and constipation in SIBO-positive pregnant women were higher than that in SIBO-negative pregnant women, and the BMI of SIBO-positive patients in the two groups was lower than that of SIBO-negative patients in each group. In addition, the TPOAb-positive rate and TSH levels were higher but the FT4 level was lower in SIBO-positive patients compared to SIBO-negative patients in study group.

Conclusion: The occurrence of subclinical hypothyroidism is related to SIBO, and the excessive growth of small intestinal bacteria may affect gastrointestinal symptoms.

Clinical Trial: http://www.chictr.org.cn/index.aspx, identifier ChiCTR1900026326.

Keywords: hypothyroidism, pregnancy, breath test, intestine, microbiota

OPEN ACCESS

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CASE REPORT

Levothyroxine Sodium Oral Solution Normalizes Thyroid Function in a Patient with Hashimoto's Disease, Gastritis, Diabetic Gastroparesis, and Small Intestinal Bacterial Overgrowth (SIBO)

Conclusion: Malabsorption of L-T4 is often seen in patients with Hashimoto's disease-related hypothyroidism and comorbid GI conditions, such as gastroparesis and SIBO. L-T4 tablets and a compounded oral suspension were inefficiently absorbed, leading to suboptimal TSH control. Switching to levothyroxine sodium oral solution resulted in sustained TSH control with subsequent resolution of symptoms. No side effects or reactions to the medication were observed, despite the patient's multiple allergies and sensitivities to food chemicals and medications.

Restless Leg Syndrome

Restless Legs Syndrome in Patients with Irritable Bowel Syndrome: Response to Small Intestinal Bacterial Overgrowth Therapy

Leonard B. Weinstock, Steve E. Fern & Stephen P. Duntley

Digestive Diseases and Sciences **53**, 1252–1256 (2008) | Cite this article

- This study suggests that SIBO associated with IBS may be a factor in some RLS patients and SIBO therapy provides long-term RLS improvement.

> Sleep Med. 2011 Jun;12(6):610-3. doi: 10.1016/j.sleep.2011.03.007. Epub 2011 May 13.

Restless legs syndrome is associated with irritable bowel syndrome and small intestinal bacterial overgrowth

> World J Gastroenterol. 2011 Oct 21;17(39):4404-7. doi: 10.3748/wjg.v17.i39.4404.

Prevalence of restless legs syndrome in patients with irritable bowel syndrome

> Neurol Res. 2018 Jul;40(7):581-585. doi: 10.1080/01616412.2018.1454704. Epub 2018 Apr 6.

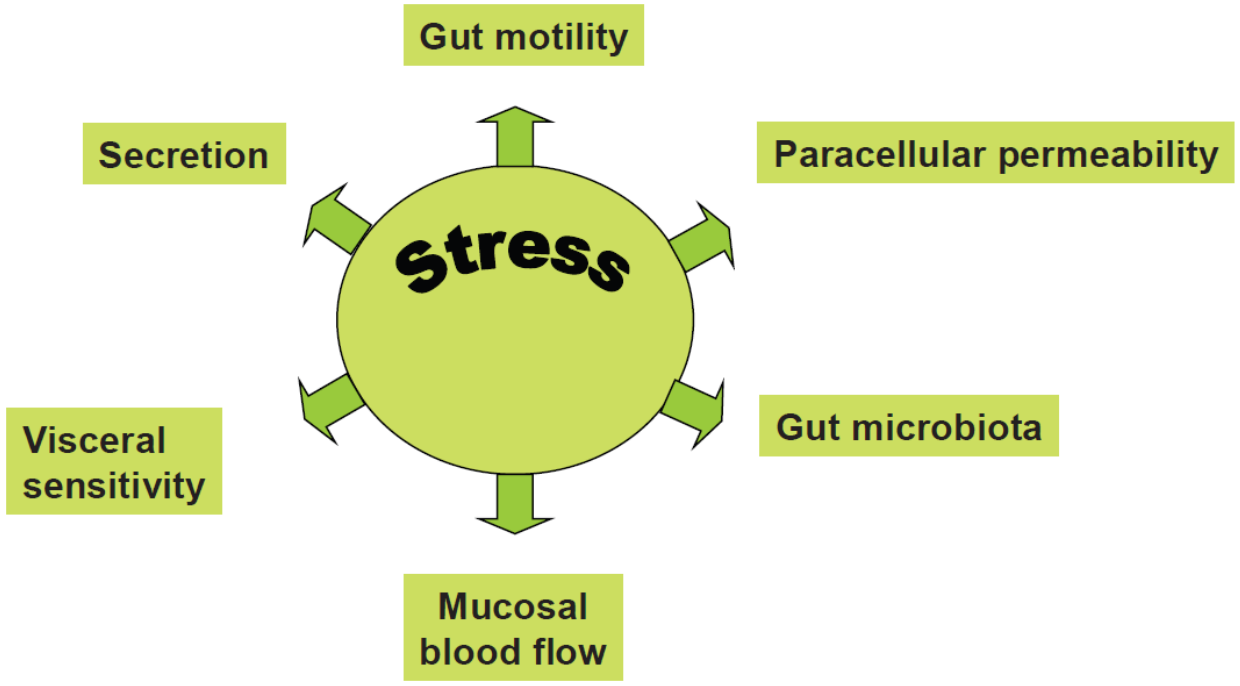
Restless legs syndrome in patients infected with Helicobacter pylori

> Inflamm Bowel Dis. 2010 Feb;16(2):275-9. doi: 10.1002/ibd.20992.

Crohn's disease is associated with restless legs syndrome

Stress-Gut Pathophysiology
(The Common Starting Point)

1. Alterations in gastrointestinal motility
2. Increase in visceral perception
3. Changes in gastrointestinal secretion
4. Increase in intestinal permeability
5. Negative effects on regenerative capacity of gastrointestinal mucosa and mucosal blood flow
6. Negative effects on intestinal microbiota



Forgotten Characterization

SIBO is characterized by abnormal bacterial colonization in the small intestine and is associated with GI symptoms such as bloating, distension, flatulence, abdominal discomfort, diarrhea, and, in severe cases, even weight loss and **significant micronutrient deficiencies.**

Clinical and Translational Gastroenterology 2023;14:e00567.

Micronutrient Deficiencies and Malnutrition

- Vitamins
  - B12, A, D, & E
- Minerals
  - Iron, calcium

Small bowel bacterial overgrowth (SBBO) syndrome is associated with excessive numbers of bacteria in the proximal small intestine. The pathology of this condition involves competition between the bacteria and the human host for ingested nutrients. This competition leads to intraluminal bacterial catabolism of nutrients, often with production of toxic metabolites and injury to the enterocyte. A complex array of clinical symptoms ensues, resulting in chronic diarrhea, steatorrhea, macrocytic anemia, weight loss, and less commonly, protein-losing enteropathy. Therapy is targeted at correction of underlying small bowel abnormalities that predispose to SBBO and appropriate antibiotic therapy. The symptoms and signs of SBBO can be reversed with this approach.

Malnutrition

Furthermore, hypochlorhydria in atrophic gastritis results in bacterial overgrowth of the stomach and small intestine, and these bacteria may bind vitamin B12 for their own use.

Annu Rev Nutr. 1999;19:357-77.

Case Reports > BMC Gastroenterol. 2020 Aug 24;20(1):283. doi: 10.1186/s12876-020-01423-8.

Small intestinal bacterial overgrowth as a cause of protracted wound healing and vitamin D deficiency in a spinal cord injured patient with a sacral pressure sore: a case report

Review > Geriatrics. 2006 Sep;61(9):21-6.

Small bowel bacterial overgrowth. An underrecognized cause of malnutrition in older adults

> Nutr Res. 2008 May;28(5):293-8. doi: 10.1016/j.nutres.2008.03.002.

Small intestinal bacterial overgrowth and thiamine deficiency after Roux-en-Y gastric bypass surgery in obese patients

Vitamin D Associated Nutrients

The patient's serum levels of calcium, phosphorous, vitamin D, and other fat-soluble vitamins also gradually increased after starting treatment for SIBO.

BMC Gastroenterol. 2020 Aug 24;20(1):283.

TREAT, ONCE

Redefining the Goal

- The primary goal of SIBO treatment is to eradicate microorganisms from the small intestine in order to reduce symptoms.
- Further treatment goals are to maintain remission, prevent possible recurrences, and correct any nutritional and vitamin deficits.

Journal of *Clinical Medicine*

MDPI

Review

How to Recognize and Treat Small Intestinal Bacterial Overgrowth?

Barbara Skrzydło-Radomańska [1] and Bożena Cukrowska [2,*]

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[2] Department of Pathomorphology, The Children Memorial Health Institute, Aleja Dzieci Polskich 20, 04-730 Warsaw, Poland
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Abstract: Small Intestinal Bacterial Overgrowth (SIBO) is a form of dysbiosis that involves increased bacterial colonization of the small intestine with some of the bacteria more characteristic of the colon microbiota. The prevalence of SIBO over recent decades has been estimated to range from 2.5 to 22% (depending on the source) and to increase with age and among individuals with comorbidities. Recently, an increase in the number of diagnosed SIBO cases has been observed, which is primarily due to the availability of noninvasive breath tests that facilitate the diagnostic process. However, SIBO is still both a diagnostic and a therapeutic problem. This review presents the pathophysiology, manifestations, diagnostics, and recommended management of SIBO.

Let Physiology Guide Treatment

Unlike the colon, the human small intestine is typically an inhospitable environment for bacteria to grow and flourish.

- Normal fasting and fed motility
- Secretion of gastric acid
- Pancreaticobiliary secretions
- Structural barriers such as the ileocecal valve
- Intact gut immune system
- Commensal bacteria

SIBO Safety Net

- Three types of protective mechanisms against SIBO
- SIBO present when overwhelmed, insufficient or absent

1. Gastrointestinal antimicrobial defense mechanisms
2. Gastrointestinal motility
3. Gastrointestinal tract anatomy

Antimicrobial Defense Mechanisms

- Connected with the production of hydrochloric acid
- Pancreatic enzyme secretion
- Bile
- Mucosal immunity
    - Insufficient immunoglobulin A secretion

J. Clin. Med. 2022, 11, 6017.

GI Motility and Anatomy

- Motility
  - Impaired small intestinal clearance
    - Absence of phase III of the migrating motor complex and retrograde peristalsis
    - Primary visceral neuropathy, myopathy
    - Secondary neuropathy (disease or drugs)
- Anatomy
  - Anatomical and surgical alterations

Small Bowel Transit Time

J Clin Gastroenterol. 2015 Aug;49(7):571-6. doi: 10.1097/MCG.0000000000000257.

Small Intestinal Transit Time Is Delayed in Small Intestinal Bacterial Overgrowth

- Patients with underlying SIBO have significant delays in SBTT as compared with those without.
- Greater than 6 hours
- Whole gut transit time slower as well

Fasting

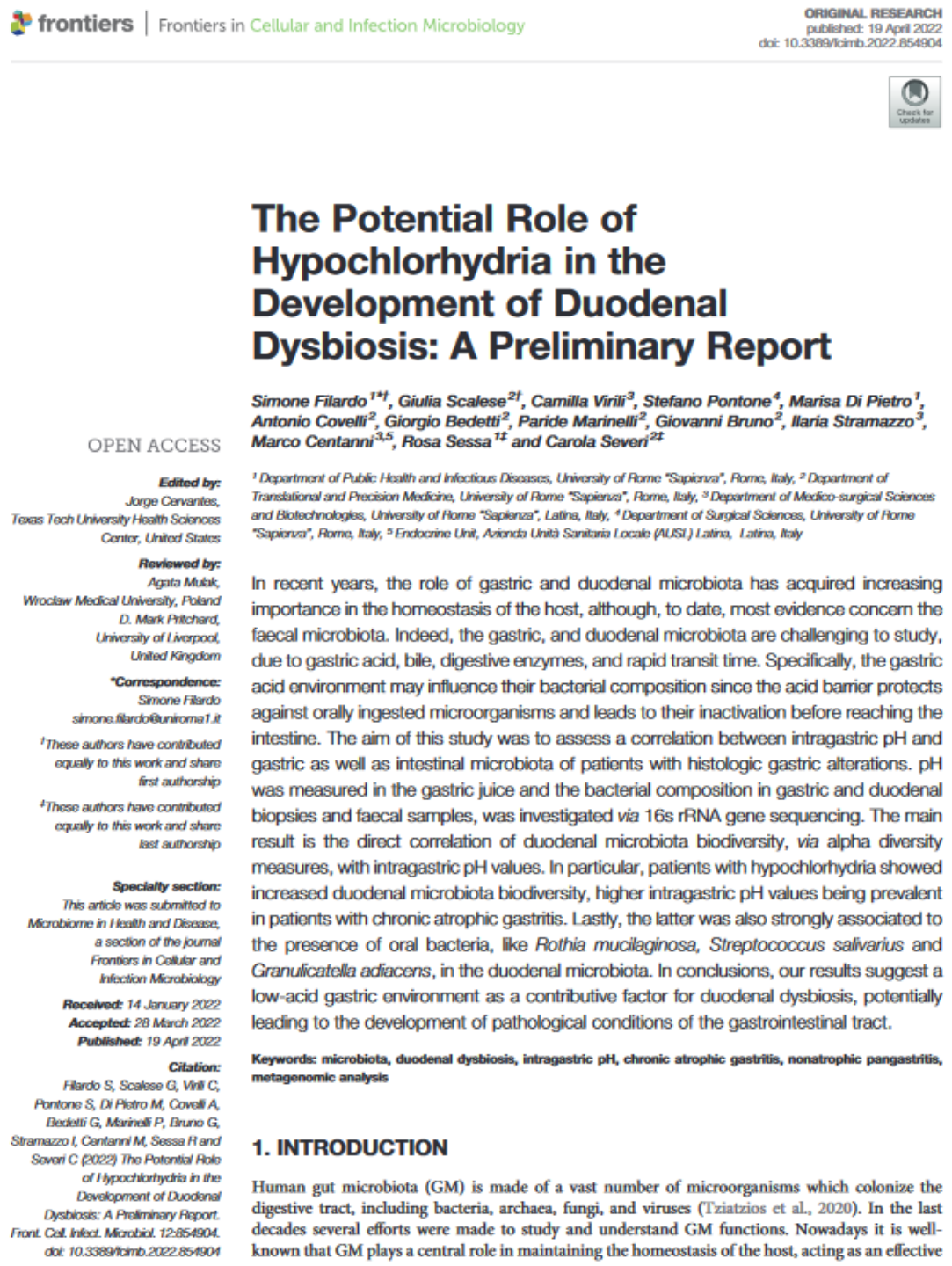
- Initiates the migratory motor complex (MMC), which is interrupted by feeding
- MMC is considered the “intestinal housekeeper”
- Band of small intestinal contractions that clear out the luminal content toward the lower intestines
- Vagal nerve stimulation enhances MMC activity in the stomach
- Serotonin stimulates duodenal MMC activity
- Absent or disordered pattern of the MMC is associated with SIBO

Am J Physiol Gastrointest Liver Physiol310: G228–G233, 2016.
Nat Rev Gastroenterol Hepatol. 2012 Mar 27;9(5):271-85.

Hypochlorhydria

In conclusions, our results suggest a low-acid gastric environment as a contributive factor for duodenal dysbiosis, potentially leading to the development of pathological conditions of the gastrointestinal tract.

Front Cell Infect Microbiol. 2022 Apr 19;12:854904.



frontiers | Frontiers in Cellular and Infection Microbiology

ORIGINAL RESEARCH
published: 19 April 2022
doi: 10.3389/fcimb.2022.854904

The Potential Role of Hypochlorhydria in the Development of Duodenal Dysbiosis: A Preliminary Report

Simone Filardo[1†], Giulia Scalese[2†], Camilla Virili[3], Stefano Pontone[4], Marisa Di Pietro[1], Antonio Covelli[2], Giorgio Bedetti[2], Paride Marinelli[2], Giovanni Bruno[2], Ilaria Stramazzo[3], Marco Centanni[3,5], Rosa Sessa[1‡] and Carola Severi[2‡]*

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In recent years, the role of gastric and duodenal microbiota has acquired increasing importance in the homeostasis of the host, although, to date, most evidence concern the faecal microbiota. Indeed, the gastric, and duodenal microbiota are challenging to study, due to gastric acid, bile, digestive enzymes, and rapid transit time. Specifically, the gastric acid environment may influence their bacterial composition since the acid barrier protects against orally ingested microorganisms and leads to their inactivation before reaching the intestine. The aim of this study was to assess a correlation between intragastric pH and gastric as well as intestinal microbiota of patients with histologic gastric alterations. pH was measured in the gastric juice and the bacterial composition in gastric and duodenal biopsies and faecal samples, was investigated *via* 16s rRNA gene sequencing. The main result is the direct correlation of duodenal microbiota biodiversity, *via* alpha diversity measures, with intragastric pH values. In particular, patients with hypochlorhydria showed increased duodenal microbiota biodiversity, higher intragastric pH values being prevalent in patients with chronic atrophic gastritis. Lastly, the latter was also strongly associated to the presence of oral bacteria, like *Rothia mucilaginosa*, *Streptococcus salivarius* and *Granulicatella adiacens*, in the duodenal microbiota. In conclusions, our results suggest a low-acid gastric environment as a contributive factor for duodenal dysbiosis, potentially leading to the development of pathological conditions of the gastrointestinal tract.

Keywords: microbiota, duodenal dysbiosis, intragastric pH, chronic atrophic gastritis, nonatrophic pangastritis, metagenomic analysis

OPEN ACCESS

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1. INTRODUCTION

Human gut microbiota (GM) is made of a vast number of microorganisms which colonize the digestive tract, including bacteria, archaea, fungi, and viruses (Tziatzios et al., 2020). In the last decades several efforts were made to study and understand GM functions. Nowadays it is well-known that GM plays a central role in maintaining the homeostasis of the host, acting as an effective

H. pylori Connection

- H. pylori-induced changes in acid secretion, in particular hypochlorhydria, may allow ingested microorganisms to survive transit through the stomach and colonize the distal intestine and colon. Such perturbation of gut microbiota, i.e. dysbiosis, may influence human health and disease.
- Acute infection – hypochlorhydria
- Chronic infection – hypo- or hyperchlorhydria
- Most patients chronically infected with H. pylori manifest a pangastritis with reduced acid secretion due to bacterial virulence factors, inflammatory cytokines, and various degrees of gastric atrophy.

Curr Top Microbiol Immunol. 2017;400:227-252.

World Journal of Gastroenterology

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World J Gastroenterol 2021 December 21; 27(47): 8040-8046

DOI: 10.3748/wjg.v27.i47.8040

ISSN 1007-9327 (print) ISSN 2219-2840 (online)

EDITORIAL

Progress in elucidating the relationship between *Helicobacter pylori* infection and intestinal diseases

Shunji Fujimori

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Abstract

Helicobacter pylori (*H. pylori*) infection causes changes to the intestinal flora, such as small intestinal bacterial overgrowth, and increases gastric acid secretion-stimulating gastrointestinal hormones, mainly gastrin, due to a decrease in gastric acid caused by atrophic gastritis. In addition, the cellular components of *H. pylori* travel through the intestinal tract, so the bacterial infection affects the immune system. Therefore, the effects of *H. pylori* infection are observed not only in the stomach and the proximal duodenum but also in the small and large intestines. In particular, meta-analyses reported that *H. pylori*-infected individuals had an increased risk of colorectal adenoma and colorectal cancer. Moreover, a recent study reported that the risk of developing colorectal cancer was increased in subjects carrying *H. pylori* vacuolating cytotoxin A antibody. In addition, it has been reported that *H. pylori* infection exacerbates the symptoms of Fabry's disease and familial Mediterranean fever attack and is involved in irritable bowel syndrome and small intestinal ulcers. On the other hand, some studies have reported that the frequency of ulcerative colitis, Crohn's disease, and celiac disease is low in *H. pylori*-infected individuals. Thus, *H. pylori* infection is considered to have various effects on the small and large intestines. However, few studies have reported on these issues, and the details of their effects have not been well elucidated. Therefore, additional studies are needed.

H. pylori

Helicobacter pylori (H. pylori) infection causes changes to the intestinal flora, such as small intestinal bacterial overgrowth, and increases gastric acid secretion-stimulating gastrointestinal hormones, mainly gastrin, due to a decrease in gastric acid caused by atrophic gastritis.

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Pancreatic Exocrine Insufficiency

- Pancreatic Enzyme Replacement Therapy (PERT) may not be ideal for all cases of SIBO at the initiation of therapy
- Possible exacerbation in some patients, but not universal
- Ironically SIBO could be the reason for poor response
- Implement post-antimicrobial therapy

Scand J Gastroenterol. 2021 May;56(5):588-593.
Pancreas. 2003 Mar;26(2):130-3.

Bile Acid Bacterial Regulation

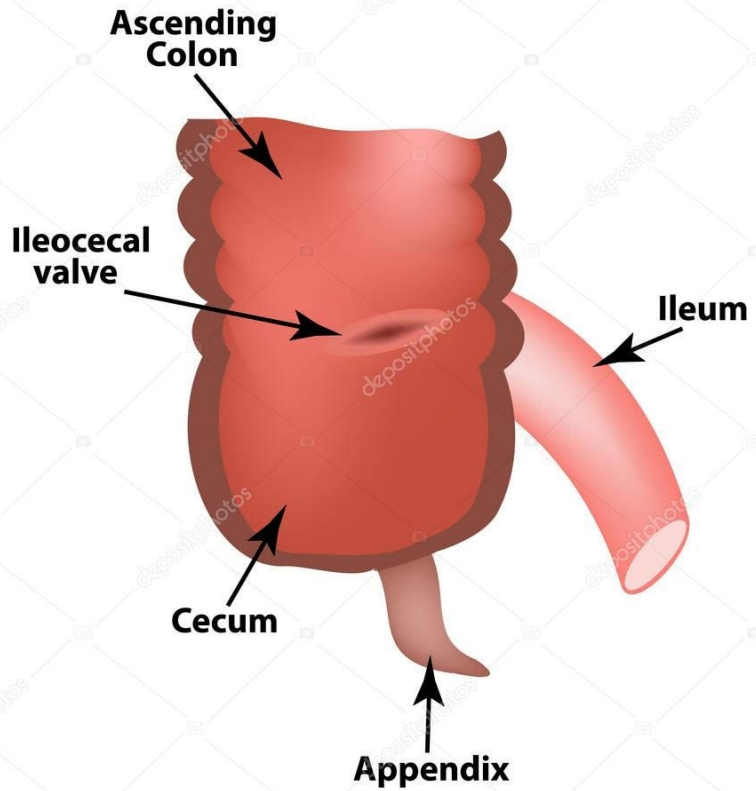
It has been demonstrated that deoxycholic acid is an order of magnitude greater than cholic acid in antibacterial activity

FEMS Microbiology Reviews. 2005;29(4):625–651.

In addition, unconjugated bile acids possess more potent antibacterial action than conjugated counterparts via membrane disruption and leakage of cellular contents

Frontiers in Microbiology. 2017;8, article 1581

Ileocecal Valve

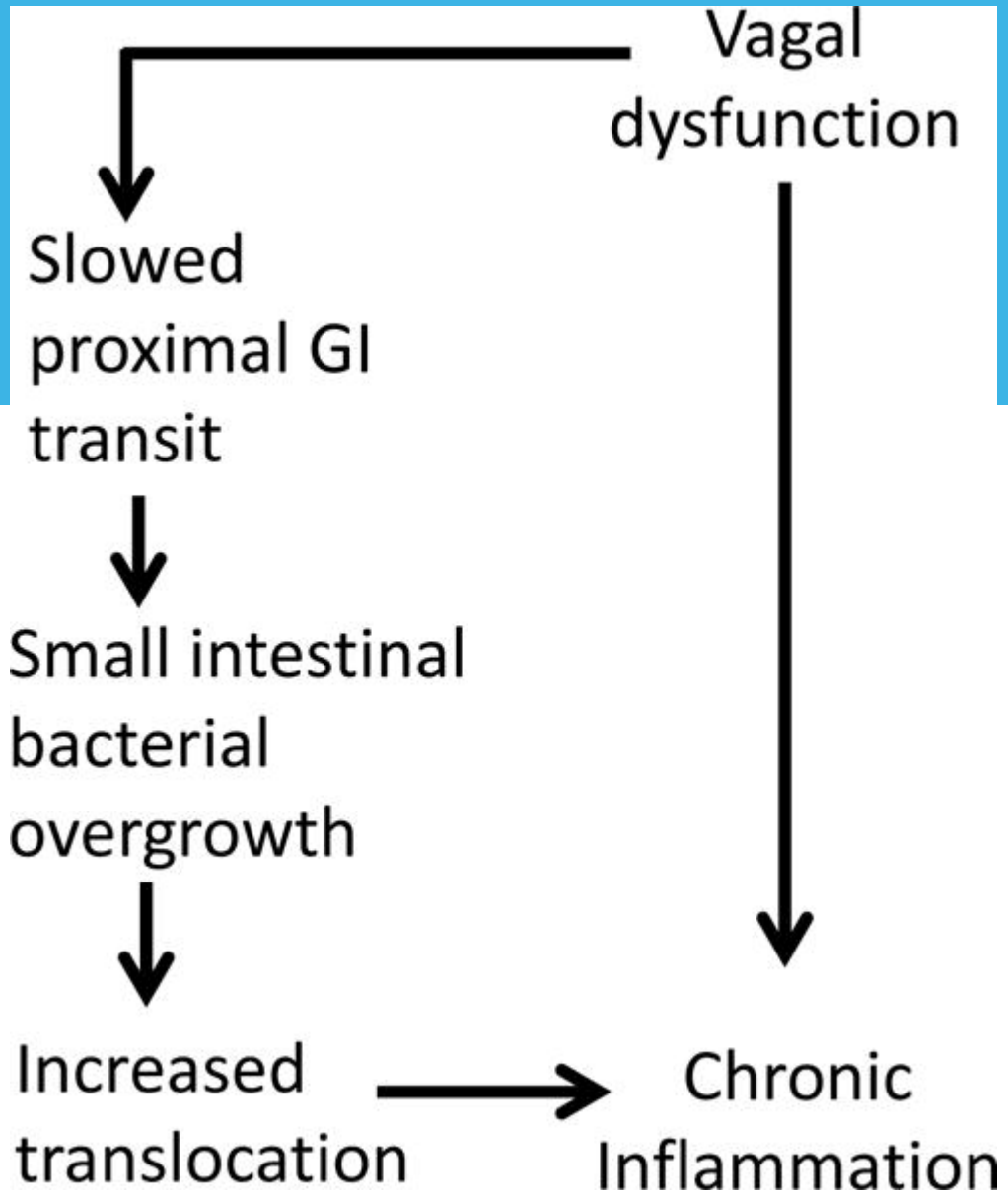


Observational Study > Dig Dis Sci. 2017 Dec;62(12):3525-3535. doi: 10.1007/s10620-017-4726-4. Epub 2017 Sep 4.

A Prospective Evaluation of Ileocecal Valve Dysfunction and Intestinal Motility Derangements in Small Intestinal Bacterial Overgrowth

Conclusions: Patients with SIBO have significantly lower ICJP, prolonged SBTT, and a higher gastrointestinal pH as compared to those without SIBO. These abnormalities may play different roles in the pathogenesis of SIBO, facilitating more targeted treatment to prevent recurrences of SIBO.

Gut-Immune Dysregulation



- SIBO associated with IL-6
- Vagal dysfunction alters cytokine networks

AIDS. 2018 Jun 1;32(9):1147-1156.

Probiotics

Review > J Clin Gastroenterol. 2017 Apr;51(4):300-311. doi: 10.1097/MCG.0000000000000814.

Probiotics for Preventing and Treating Small Intestinal Bacterial Overgrowth: A Meta-Analysis and Systematic Review of Current Evidence

Therefore, the present findings indicated that probiotics supplementation could effectively decontaminate SIBO, decrease H2 concentration, and relieve abdominal pain, but were ineffective in preventing SIBO.

Probiotics vs. Antibiotics

Results: Thirteen (52%) subjects receiving metronidazol and 20 (82%) receiving the probiotic referred clinical improvement after the treatment. A statistically significant difference favoured the use of the probiotic (P = 0.036). All the study patients completed treatment.

(Lactobacillus casei (3.3 x 10(7) UFC), Lactobacillus plantarum (3.3 x 10(7) UFC), Streptococcus faecalis (3.3 x 10(7) UFC) and Bifidobacterium brevis (1.0 x 10(6) UFC for 5 days)

Conclusions: Based on this pilot study results, we can suggest that the probiotic herein used has a higher efficacy than metronidazol in the early clinical response of patients with chronic abdominal distension and SIBO.

BIOTICS RESEARCH®

Acta Gastroenterol Latinoam. 2010 Dec;40(4):323-7.

Probiotics – Choose Wisely

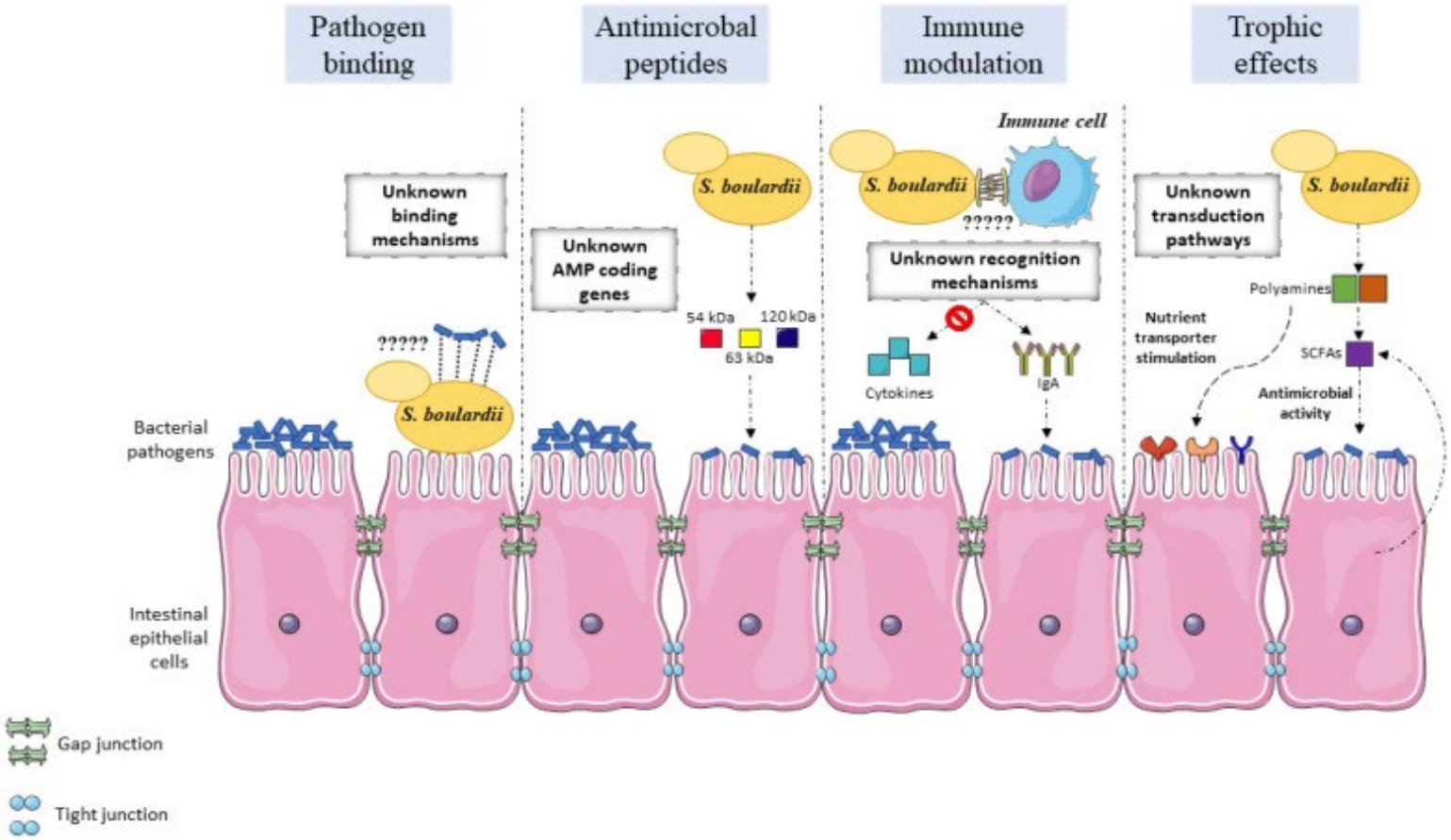
- Probiotic administration in SIBO patients resulted in exacerbated bloating, flatulence, metabolic lactic acidosis, and “brain fog,” with these symptoms subsiding after the probiotic preparation was discontinued and antibiotic therapy was initiated.
- The effects of probiotic therapy in SIBO are strongly strain-dependent, and not all probiotics are equally effective.

BIOTICS RESEARCH®

J. Clin. Med. 2022, 11, 6017.

Saccharomyces Boulardii

- Monotherapy with Saccharomyces boulardii was more effective than metronidazole to eradicate SIBO
- Best results noted by combining the two therapies



J Fungi (Basel). 2020 Jun 4;6(2):78.
Dig. Dis. Sci. 2020, 65, 1134–1143.

One vs. Many

Conclusion: In this study, the treatment of small intestinal bacterial overgrowth using rotating antibiotics was more effective than treatment using a single course of antibiotic. Remission was associated with improvement in both quality of life and bloating.

ORIGINAL ARTICLE

ueg journal WILEY

The effectiveness of rotating versus single course antibiotics for small intestinal bacterial overgrowth

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Abstract

Background: Small intestinal bacterial overgrowth treatment is usually based on antibiotics with no guidelines available.

Objective: This study aimed to investigate the efficacy of different antibiotics to treat small intestinal bacterial overgrowth.

Methods: Consecutive patients referred to our tertiary center and diagnosed with intestinal bacterial overgrowth were retrospectively included. Patients were diagnosed using a 75 g glucose breath test. Patients were treated either with a single antibiotic (quinolone or azole) or rotating antibiotics (quinolone and azole, one after the other) for 10 consecutive days per month for 3 months. A negative glucose breath test after antibiotic treatment was considered as remission. Quality of life (GIQLI) and gastrointestinal severity (IBS-SSS) were assessed before and after antibiotic treatment. Symptomatic evaluation was realized in simple blind of glucose breath test result: patients were unaware of their results.

Results: Between August 2005 and February 2020, 223 patients were included in the analysis (female 79.8%, mean age 50.2 ± 15.7 years). Remission was observed in 119 patients (53.4%) after one course of antibiotics and was more frequent in patients receiving rotating antibiotics than in patients receiving a single antibiotic (70.0% vs. 50.8%, $p = 0.050$). Remission was associated with a significant improvement in quality of life ($p = 0.035$) and in bloating ($p = 0.004$).

Conclusion: In this study, the treatment of small intestinal bacterial overgrowth using rotating antibiotics was more effective than treatment using a single course of antibiotic. Remission was associated with improvement in both quality of life and bloating.

United European Gastroenterol J. 2021 Jul;9(6):645-654.

Existing Treatment Challenges

- Antibiotic therapy (Rifaxamin, Metronidazole / Flagyl)
  - *Side effects (gut microbiome disruption, risk of C.diff)*
  - *High recurrence rate*
  - *Multiple rounds necessary*
- Nutrition support
  - *Variations of effectiveness*
  - *Many are not validated*
  - *Have not been clinically studied*
- Lifestyle changes – *compliance*
- Diet modification – *overly restrictive*

Breakthrough SIBO Study

Gerard Mullin's study highlights

- 4-week clinical trial, over 100 participants
- Patients received either Rifaximin or herbal therapy

Normal breath test after 30 days

- **46%** herbal therapy vs **34%** Rifaximin
- 1.85-fold greater likelihood of improvement with herbal therapy

ORIGINAL RESEARCH

Herbal Therapy Is Equivalent to Rifaximin for the Treatment of Small Intestinal Bacterial Overgrowth

Victor Chedid, MD, *United States*; Sameer Dhalla, MD, *United States*; John O. Clarke, MD, *United States*; Bani Chander Roland, MD, *United States*; Kerry B. Dunbar, MD, *United States*; Joyce Koh, MD, *United States*; Edmundo Justino, MD, *United States*; Eric Tomakin, RN, *United States*; Gerard E. Mullin, MD, *United States*

FC Cidal	Dysbiocide
Proprietary blend - 500 mg: 1 capsule	Proprietary Blend 950 mg per 2 capsules

Conclusion: SIBO is widely prevalent in a tertiary referral gastroenterology practice. Herbal therapies are at least as effective as rifaximin for resolution of SIBO by LBT. Herbals also appear to be as effective as triple antibiotic therapy for SIBO rescue therapy for rifaximin non-responders. Further, prospective studies are needed to validate these findings and explore additional alternative therapies in patients with refractory SIBO.

Glob Adv Health Med . 2014 May;3(3):16-24.

BIOTICS RESEARCH®

46% Botanicals vs 34% Antibiotics

WHAT WAS IN THE STUDY:

- 2 FC-Cidal™ BID
- 2 Dysbiocide® BID

46% & Beyond

History has validated an updated protocol shown to be even more effective

WHAT IS IN THE KIT:

- 2 FC-Cidal™ TID
- 2 Dysbiocide® TID
- 2 A.D.P.® TID

Plus patient-friendly guidebook

Three of Biotics Research's Most Trusted Antimicrobial Formulas*

*This statement has not been evaluated by the Food and Drug Administration. This product is not intended to diagnose, treat, cure, or prevent any disease.

Introducing **SBO Kit**

*A 30-day intensive gut support program designed to restore microbial balance**

COMPONENTS:

A.D.P.® - Patented formula of sustained release emulsified oil of oregano

FC-Cidal™ - Proprietary blend of herbs that support healthy GI function (French Tarragon, Indian Tinospora, Horsetail, Thyme, Pau D'Arco, Stinging Nettle, Olive)

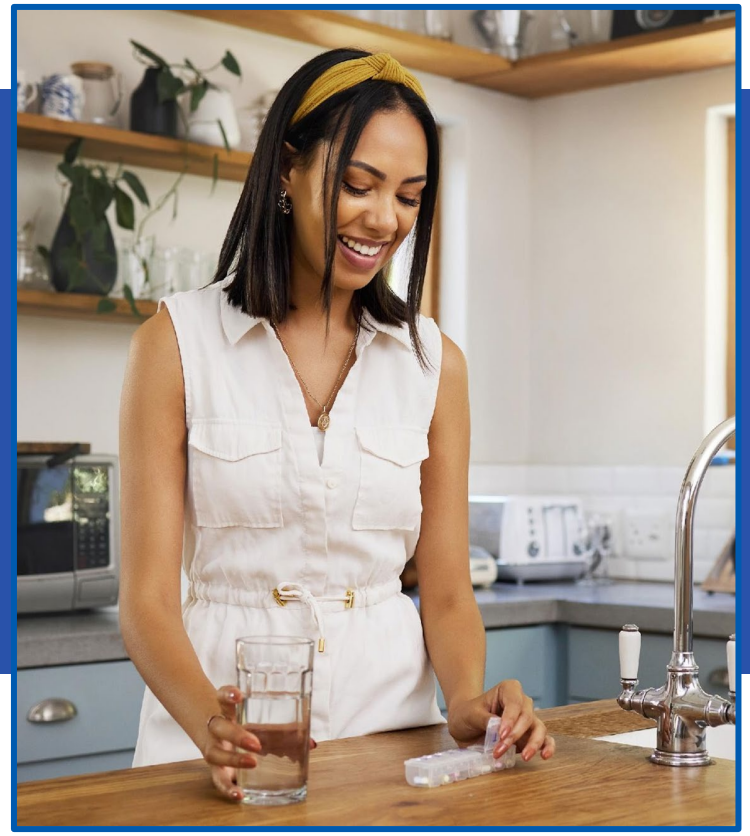
Dysbiocide® - Synergistic herbal formula supporting healthy gut microbiota (Dill, Stemona, Wormwood, Java Brucea, Chinese Pulsatilla, Jamaica Quassia, Hedyotis, Cutch Tree, Yarrow)

Patient Guidebook - Step-by-step guide to support patients on the program

*This statement has not been evaluated by the Food and Drug Administration. This product is not intended to diagnose, treat, cure, or prevent any disease.

SBO Kit: 30-Day Program

Easy-to-follow 3- Pronged Patient Program



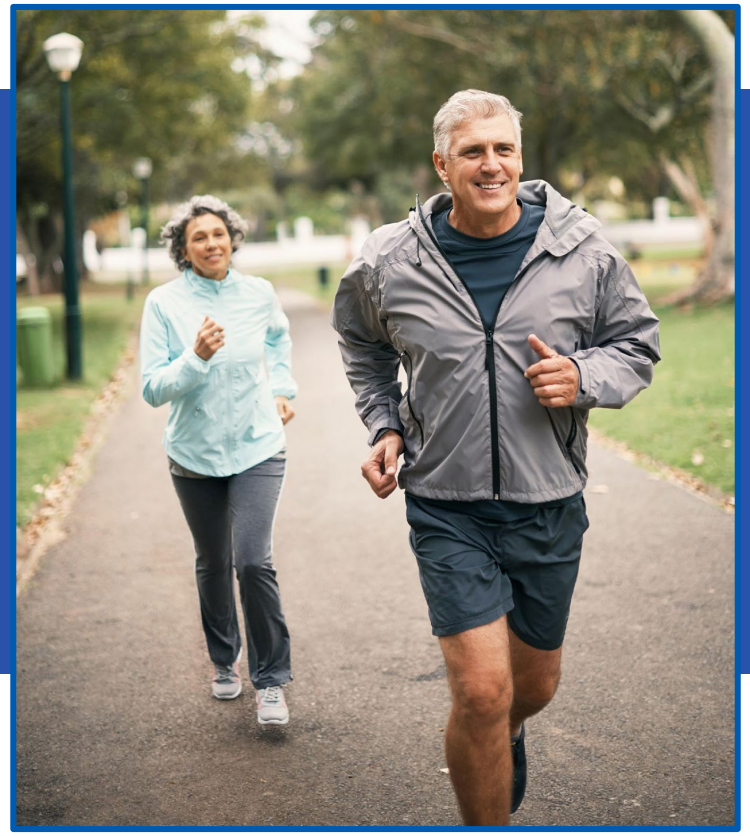
Supplements:

A.D.P.®, FC-Cidal™, Dysbiocide®

For all products, take 2 capsules, 3 times per day away from food

Diet:

Eat a whole foods diet of unprocessed foods and scale with restriction as necessary (foundation) and other dietary suggestions continue if necessary.



Lifestyle:

Stress management
Regular exercise
Avoid smoking and excessive alcohol consumption

Diet, Supplement & Lifestyle Changes

Five Key Dietary Principles

1. **Eat a clean whole foods diet:** The main key to obtaining microbial balance is to eat a diet of clean and unprocessed foods rich in essential nutrients.
2. **Include phytonutrient-rich foods:** Fresh, organic leafy green vegetables, berries, nuts and fatty fish help support healthy inflammatory pathways and add important nutrients to promote bacterial balance.
3. **Avoid sugar:** Sugar feeds harmful gut bacteria so limiting or eliminating sugar altogether is recommended. Natural sugars such as those found in whole fruits and yogurts are acceptable, but even excessive natural sugars are not recommended.
4. **Eat less frequent meals:** Eating less frequent meals promotes motility towards the large intestine, clearing the contents of the small intestines, including microbes. Three (3) meals a day with no snacks will create optimal digestion.
5. **Add fermented foods:** Fermented foods such as sauerkraut, kimchi and plain yogurt provide probiotics that promote the growth of beneficial bacteria and support digestive health.

NOTE: Ask your healthcare practitioner about a low FODMAP diet, which may prove beneficial to some patients. A low FODMAP (fermentable oligo-, di-, mono- saccharides and polyols) diet is defined as avoiding certain types of carbohydrates that can be difficult to digest. These carbohydrates, such as garlic, onions, wheat, legumes and certain fruits, also potentially feed the unwanted bacteria in the small intestine.

These five principles are general guidelines; always check with your healthcare provider for individual recommendations.

BIOTICS RESEARCH®

SBO KIT

INTENSIVE GUT MICROBIAL SUPPORT

30-Day Program

Guidebook

Dietary Modifications

STANFORD
HOSPITAL & CLINICS
Stanford University Medical Center

Digestive Health Center
Nutrition Services

The Low FODMAP Diet
(FODMAP=Fermentable Oligo-Di-Monosaccharides and Polyols)

FODMAPs are carbohydrates (sugars) that are found in foods. Not all carbohydrates are considered FODMAPs.

Dietary Controversies

- It is worth emphasizing that the period of complete elimination of FODMAP from the diet of SIBO patients should not exceed six weeks; and, if ineffective, this diet should not be used again in the future.
- There is no evidence supporting the use of a gluten-free diet in the treatment of either SIBO or IBS.
- The use of elemental diets, which contain pre-digested nutrients, is not recommended in SIBO despite some promising study reports.

J. Clin. Med. 2022, 11, 6017.

Dairy

Vopr Pitan. 2022;91(2):15-20. doi: 10.33029/0042-8833-2022-91-2-15-20. Epub 2022 Mar 14.

[Assessment of the state of the small intestine microbiota in children on a long-term dairy-free diet]

[Article in Russian]

A V Nalyotov [1 2], N A Svistunova [1]

- **Conclusion**. Thus, among children of primary school age who follow a long-term dairy free diet, SIBO is significantly more often recorded relative to children who are on a traditional type of diet.
- 55.0% vs 20.0%

Post Infectious SIBO

- Glutamine as a primary intervention – **5gm tid** po 8 weeks
- Glutamine
  - Essential amino acid in humans
  - Major energy source for rapidly dividing epithelial cells of the gastrointestinal tract
  - Depleted during infection or illness
  - Leads to atrophy of intestinal epithelial cells and intestinal hyperpermeability
  - Supplementation of the diet with glutamine can restore normal intestinal permeability
  - Decrease bacterial and toxin translocation after intestinal injury

Gut. 2019 June ; 68(6): 996–1002.

Complete SIBO Treatment Plan

1. Treatment (1 month)
    - Listen for SIBO symptom sequela
    - Test as needed
    - Recommend SBO Kit
2. Prevent Recurrences (9 months)
    - Identify and correct issues with motility and GI secretions
3. Correct nutrient deficiencies (6 months)
    - Use testing to guide recommendations as needed

BIOTICS RESEARCH®

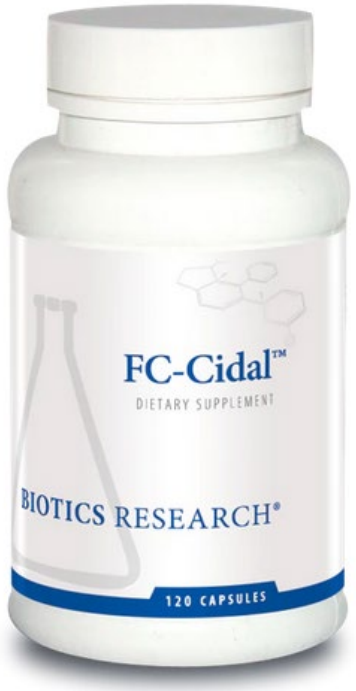
SBO Kit

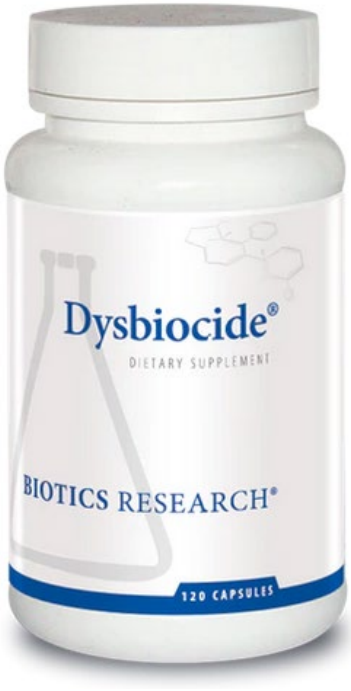
HIGHLIGHTS

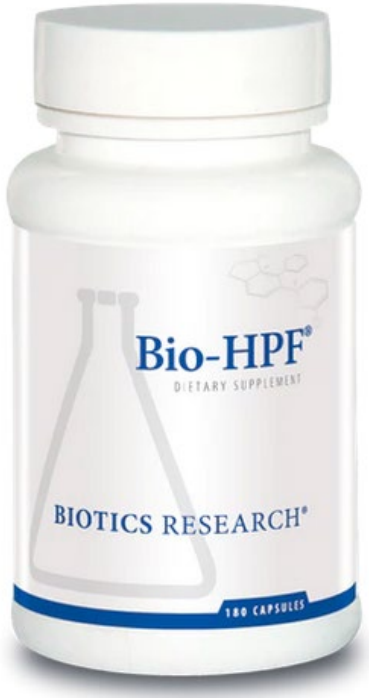
- 30-day program
- Targeted antimicrobial therapy
- Most trusted botanical formulas
- Easy-to-follow guidebook
- Dietary guidelines
- Lifestyle suggestions
- Patient compliance

For the “One Percenters”

Antimicrobials



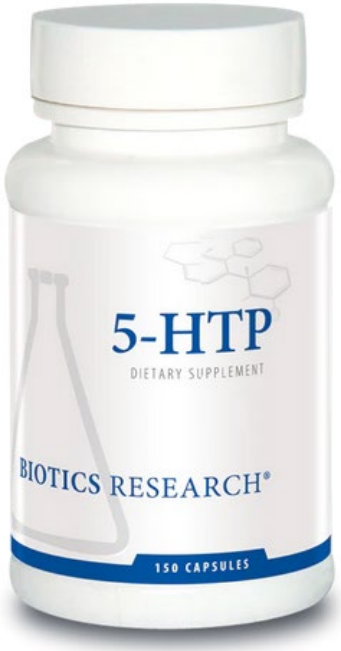




Treatment Summary

Small Intestinal Motility

Vagal Nerve Stimulation



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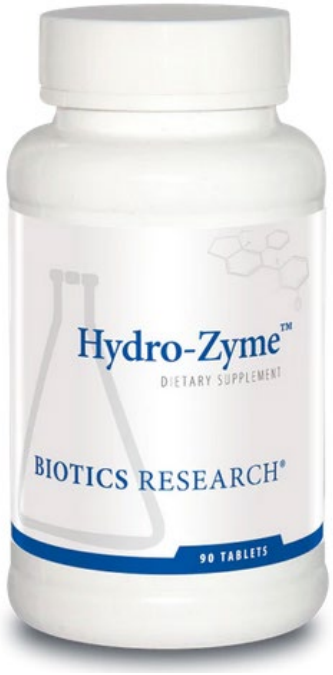
Treatment Summary

Gastric Acid Replacement



Pancreaticobiliary Support

Motility-Zyme™

Functional Approach to Constipation

- Non-laxative formula designed to improve gut function, providing a long-term solution versus temporary or habit-forming “fix” with products used in purging, agitating, catharsis for motility

1. **Ginger** – gut nervous system enhancement
2. **Artichoke** – bowel calming
3. **5-HTP** – supports motility
4. **Magnesium Citrate** – bowel relaxation

BIOTICS RESEARCH®

Treatment Summary

Ileocecal Valve Support

- Ileocecal Valve Massage
- Vagal Nerve Support

Commensal Bacteria

BIOTICS RESEARCH®

Inability to Resolve Causes

- Some patients have persistent relapse due to chronic dysfunction (disease, surgical, anatomical)
- Remain persistent with digestive support
- Rotation of low dose antimicrobials
- Low dose probiotics as a consideration
    - Pulsed

Take Aways

- Is SIBO the cause or the result of the associated condition?
- Consider the bidirectional relationship of SIBO associated conditions.
- Understand the limitations of SIBO diagnostic testing and how to correlate to other findings.
- Implementing antimicrobial support is only the first phase.
- Follow up with correcting underlying causes and nutrient deficiencies.

Insufficiency Dysbiosis

COMMENSAL/KEYSTONE BACTERIA

COMMENSAL BACTERIA	Result	Reference
Bacteroides fragilis	3.29e9	1.6e9 - 2.5e11
Bifidobacterium spp.	4.18e8	> 6.7e7
Enterococcus spp.	1.36e6	1.9e5 - 2.0e8
Escherichia spp.	<dl L	3.7e6 - 3.8e9
Lactobacillus spp.	1.47e5 L	8.6e5 - 6.2e8
Enterobacter spp.	1.93e6	1.0e6 - 5.0e7
Akkermansia muciniphila	2.52e4	1.0e1 - 8.2e6
Faecalibacterium prausnitzii	5.90e2 L	1.0e3 - 5.0e8
Roseburia spp.	4.56e7 L	5.0e7 - 2.0e10
BACTERIAL PHYLA		
Bacteroidetes	3.47e11 L	8.6e11 - 3.3e12
Firmicutes	1.14e10 L	5.7e10 - 3.0e11
Firmicutes:Bacteroidetes Ratio	0.03	< 1.0

Gut Microbiota and Healthy Aging

Age-related microbial dysbiosis

Reshaping of immune responses

Immunosenescence (insufficiency) and inflammaging (over-reaction) manifest

Linked to age associated enteric and extraenteric diseases

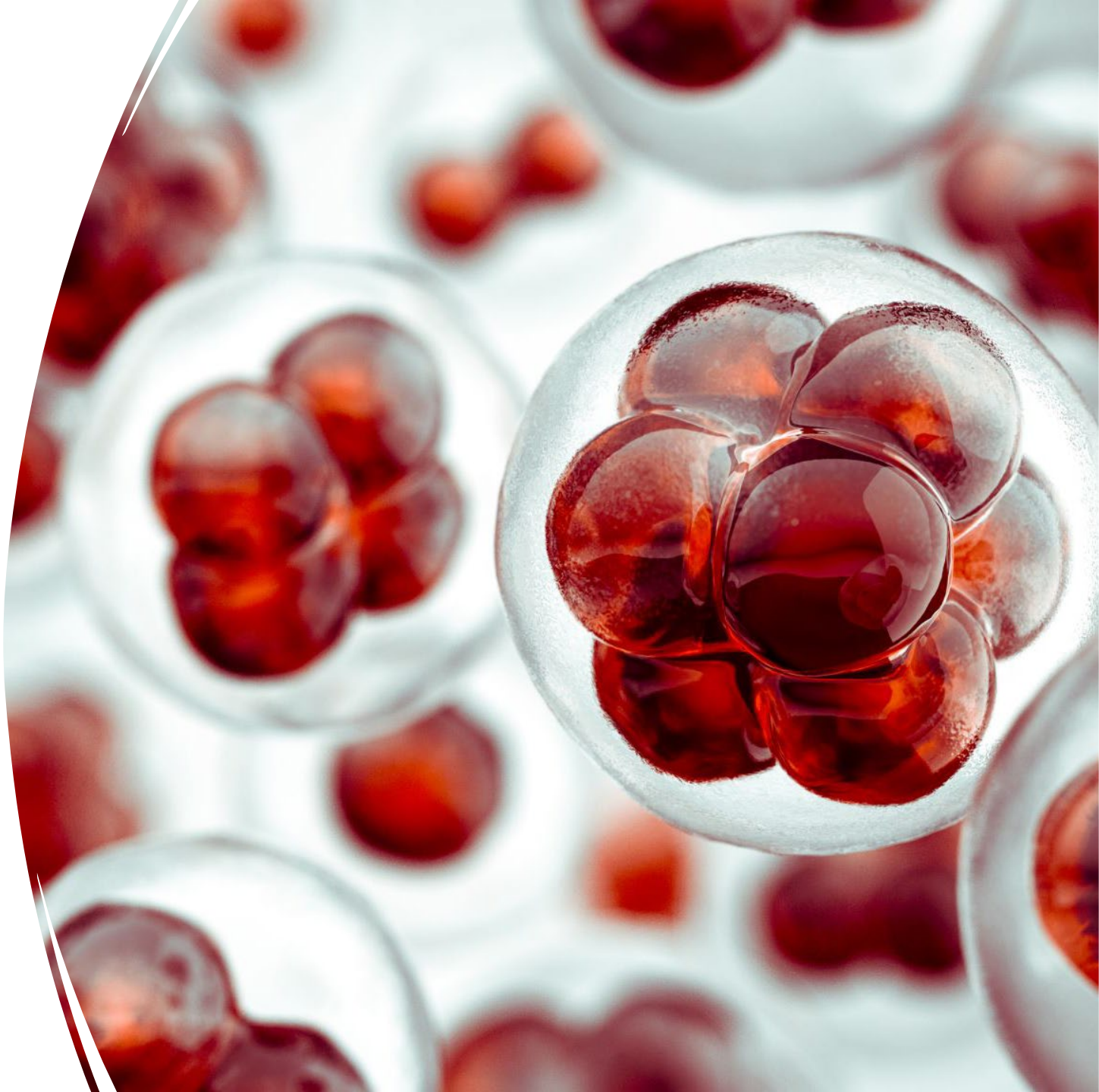
- Colorectal cancer, sarcopenia and physical frailty
- Nonalcoholic fatty liver disease(NAFLD), coronary heart disease
- Neurodegenerative diseases, Type 2 diabetes mellitus

Microbiota becomes a target for aging

Age-Related Dysbiosis

Gut Frailty

- Stomach, small intestine and large intestine
- Inflammation related immune cells
- Natural killer cells
- Macrophages
- Chronic inflammation



Gut Frailty

Indicators of Frailty (3 or more required)
Unintentional weight loss of 4.5 kg or more per year or a weight loss of 5% or more
Self-reported exhaustion occurring on 3–4 days or more per week.
Slow walking speed
Decreased grip strength
Decreased physical activity

Indicators of Gut Frailty

1. Epigastric pain and discomfort
2. Bowel irregularities such as constipation or diarrhea
3. Abdominal pain and bloating
4. Stress-related symptoms
5. Decreased appetite and weight loss

Digestion 2023;105:49–57

Izumo Scale (Consistent / Reproducible)

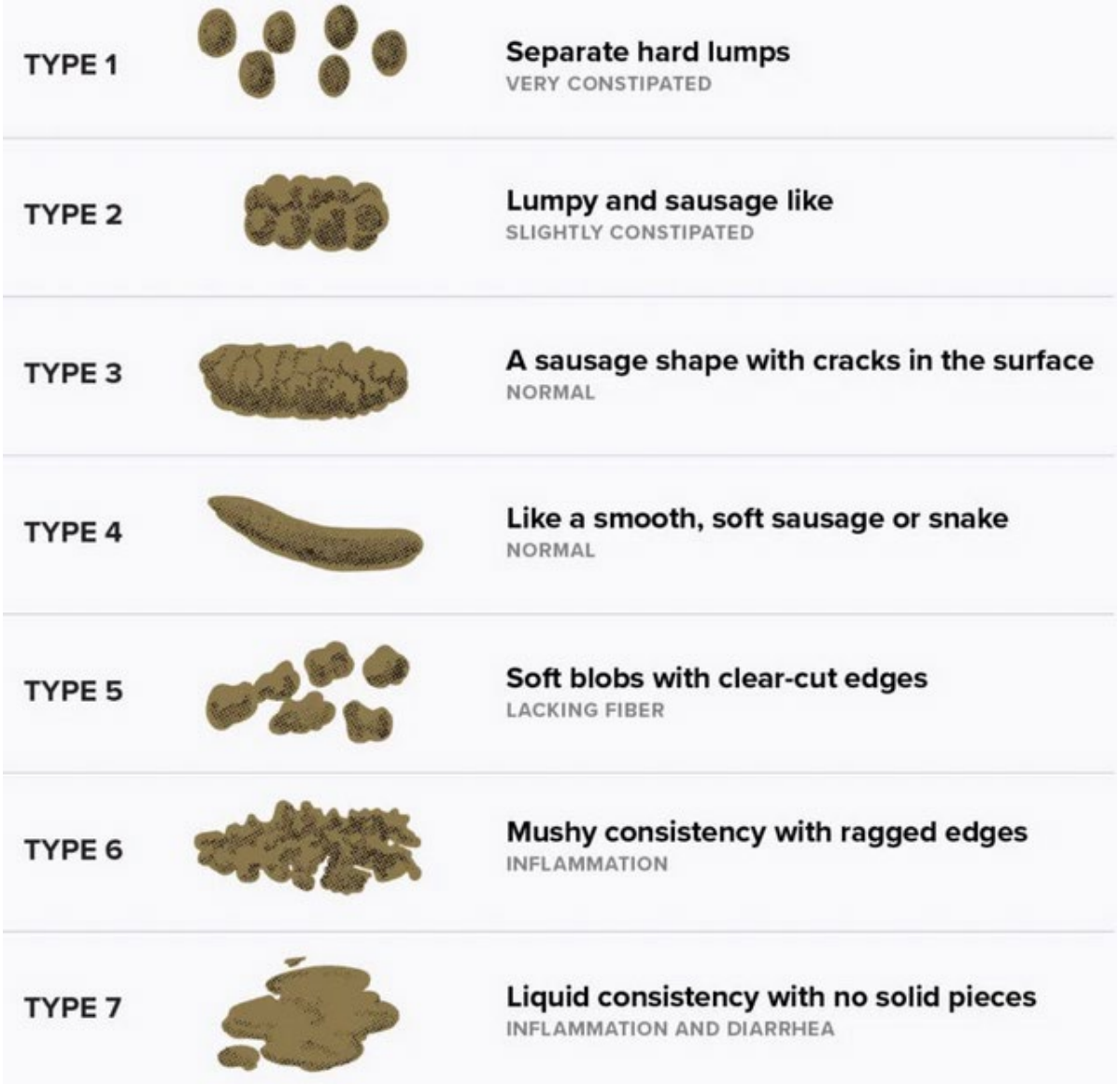
Question	Not bothered	Not so bothered	Slightly bothered	Bothered	Strongly bothered	Intolerably bothered
1. Are you bothered by acid reflux?	0	1	2	3	4	5
2. Are you bothered by heartburn centered in the anterior chest?	0	1	2	3	4	5
3. Are you bothered by throat discomfort?	0	1	2	3	4	5
4. Are you bothered by epigastric pain?	0	1	2	3	4	5
5. Are you bothered by hunger epigastric pain?	0	1	2	3	4	5
6. Are you bothered by epigastric burning sensation?	0	1	2	3	4	5
7. Are you bothered by early satiation?	0	1	2	3	4	5
8. Are you bothered by post-prandial long-lasting epigastric fullness or nausea?	0	1	2	3	4	5
9. Are you bothered by epigastric bloating?	0	1	2	3	4	5
10. Are you bothered by a feeling of incomplete defecation?	0	1	2	3	4	5
11. Are you bothered by constipation or hard stools?	0	1	2	3	4	5
12. Are you bothered by stress-related constipation?	0	1	2	3	4	5
13. Are you bothered by fecal urgency?	0	1	2	3	4	5
14. Are you bothered by diarrhea or soft stools?	0	1	2	3	4	5
15. Are you bothered by stress-related diarrhea?	0	1	2	3	4	5

Izumo Scale Interpretation

- 15 questions in 5 domains
  1. Reflux – questions 1-3
  2. Pain – questions 4-6
  3. Fullness – questions 7-9
  4. Constipation – questions 10-12
  5. Diarrhea – questions 13-15
- Scale of 0-75
- Closer to 75, greater the level of dysfunction

Bristol Stool Form

- Abnormal bowel movements
  - Type 1, 2, 6 or 7
- Additional definitions
  - Bowel frequency >3/day
  - Bowel frequency <3/week



Constipation

- Key symptom of gut frailty
- Correlated with significantly lower survival rates
- Disease associations
  - Chronic kidney disease, Cardiovascular diseases, and neurodegenerative disorders like Parkinson's disease
- At least 1 in 8 patients
- However, patients significantly underrecognize constipation
- More than discomfort
- Not simply a lack of fiber
- Neurogenic and microbials causes are most common

Possible Drivers of Gut Frailty

- Decrease in mucin production
  - Medications – ex. Aspirin
  - Reduction in dietary fiber
  - Reduction of goblet cells secondary to permeability and inflammation
- Decreased tight junction function
- Decreased peristalsis
- Decreased cell turnover
- Stem cell dysfunction
- Increased senescent cells
- Insufficiency of the microcirculation
- Dysbiosis of gut microbiota

Gut Dysbiosis and Frailty

- Decline of Faecalibacterium prausnitzii
- α-diversity is NOT associated with mortality
- Mortality strongly associated with Enterobacteriaceae family
  - Dominant in Proteobacteria
  - Facultative anaerobes
    - Escherichia
    - Klebsiella
    - Proteus
    - Salmonella
    - Shigella
    - Yersinia

Prevention and Therapy of Gut Frailty

- Diets/food factors – dietary fiber, secondary bile acids
- Drugs/senolytics
- Fecal microbiota transplantation
- Probiotics/prebiotics – Akkermansia mucinophilia
- Metabolites of gut microbiota (postbiotics) - butyrate
- Physical activity
  - Increased levels of bile acids
  - Benefit mediated through the gut microbiota

Tributyrin - Postbiotic

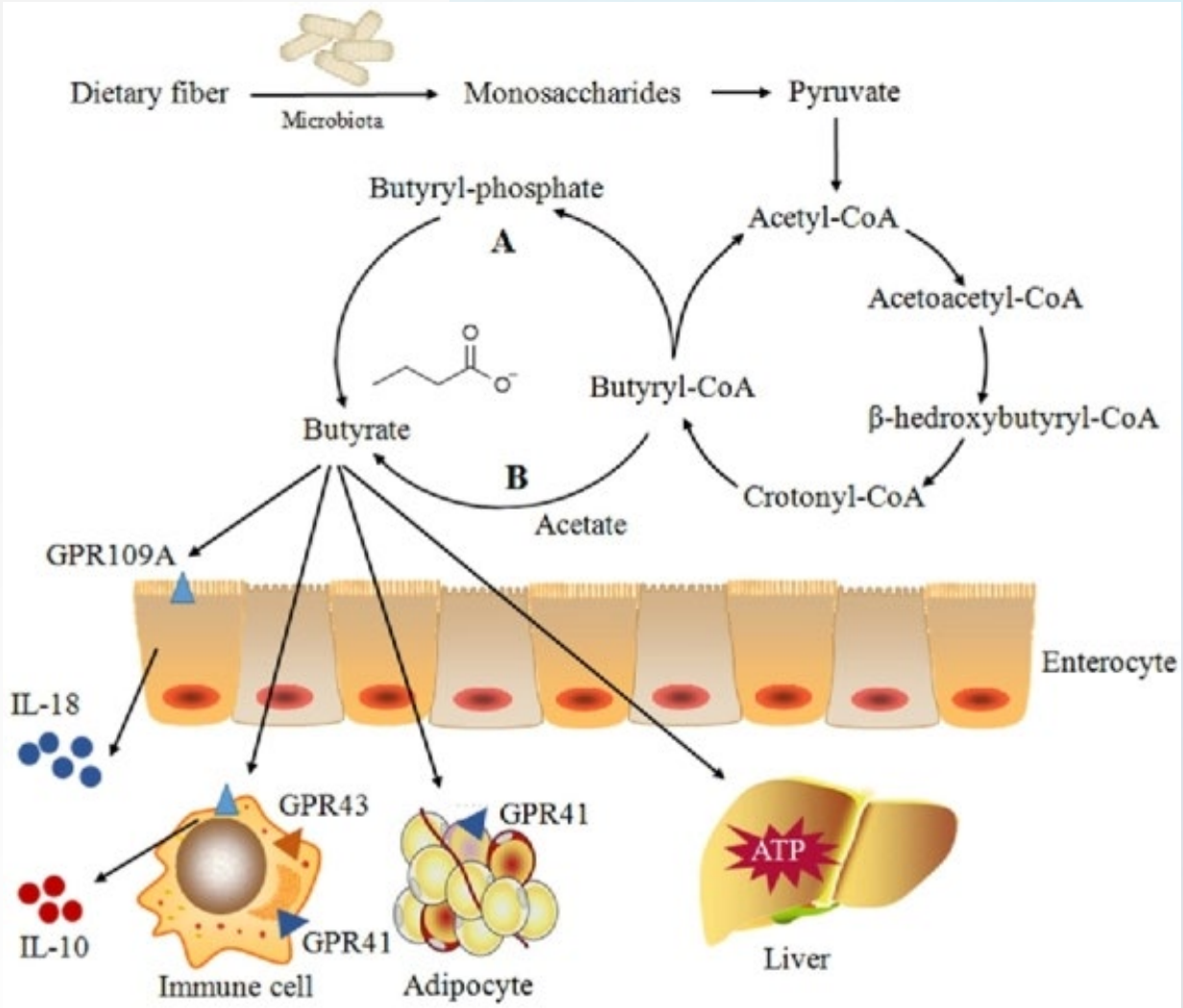
- Butyric acid prodrug
- Reduces rapid uncontrolled cellular growth
- Enhances differentiation
- Upregulates vitamin D receptors

- J Nutr. 2001 Jun;131(6):1839-43.

Natural Butyrate

Production

- Short-chain fatty acid (SCFA)
- Produced by commensal bacteria
- Produced in the colon
- Derived from the fermentation of dietary fiber
    - Primarily indigestible plant polysaccharides and resistant starches.



Natural Butyrate

Production

In addition to its well-recognized role as the preferred energy source for colonocytes, butyrate has now been shown to have a much broader physiological role that extends beyond the colon, ***influencing systemic metabolic and immune function***, as well as reducing ***intestinal permeability and inflammation***.

nutrients

MDPI

Review

Short Chain Fatty Acids in the Colon and Peripheral Tissues: A Focus on Butyrate, Colon Cancer, Obesity and Insulin Resistance

Sean M. McNabney and Tara M. Henagan *

Department of Nutrition Science, Purdue University, West Lafayette, IN 47907, USA; smcnabne@purdue.edu
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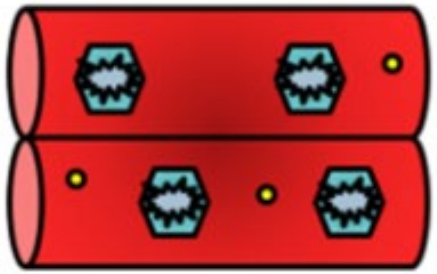
Received: 19 October 2017; Accepted: 5 December 2017; Published: 12 December 2017

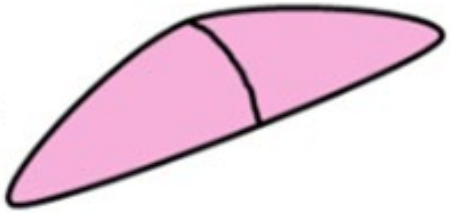
Abstract: Increased dietary fiber consumption has been associated with many beneficial effects, including amelioration of obesity and insulin resistance. These effects may be due to the increased production of short chain fatty acids, including propionate, acetate and butyrate, during fermentation of the dietary fiber in the colon. Indeed, oral and dietary supplementation of butyrate alone has been shown to prevent high fat-diet induced obesity and insulin resistance. This review focuses on sources of short chain fatty acids, with emphasis on sources of butyrate, mechanisms of fiber and butyrate metabolism in the gut and its protective effects on colon cancer and the peripheral effects of butyrate supplementation in peripheral tissues in the prevention and reversal of obesity and insulin resistance.



Colon:
Primary energy source for colonocytes
Reduced inflammatory cytokines
Maintenance of tight junctions
Regulation of colonic lumen pH
Enhanced Na+ absorption
Promotes cell cycle arrest and apoptosis in colon cancers
Increased production and secretion of satiety hormones
Altered gut microbiota

Skeletal Muscle:
Increased lean body mass
Increased and more complete β-oxidation
Increased capacity for substrate switching
Increased oxidative, type 1 fiber percentage
Decreased lipid accumulation

Butyrate

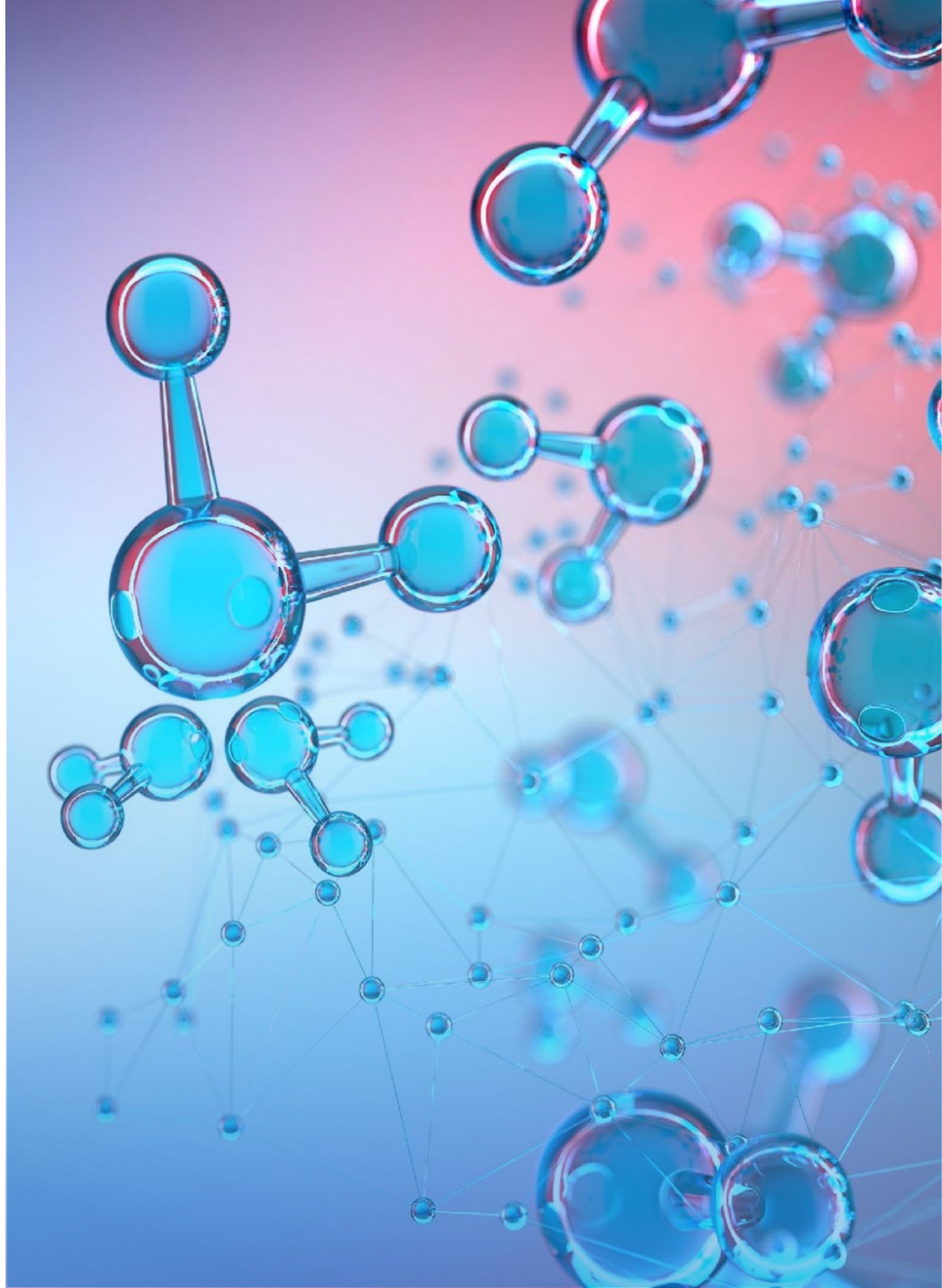


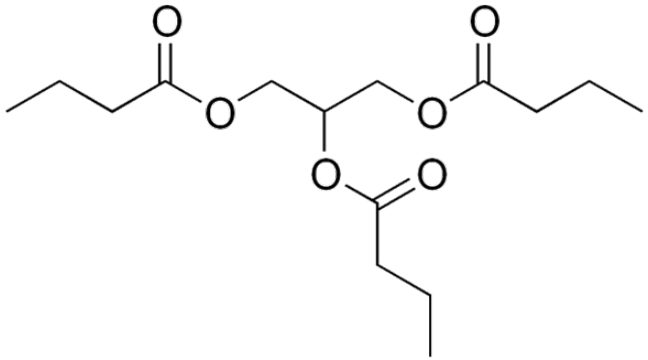
Liver:
Reduced inflammatory mediators
Enhanced antioxidant systems
Increased β-oxidation
Reduced lipid deposition

Adipose Tissue:
Decreased adipose tissue mass
Increased lipolysis *in vitro*
Increased β-oxidation
Increased SNS outflow and β-adrenergic stimulation on adipocytes
Increased leptin production and secretion
Decreased inflammation
Browning of white adipose tissue
Enhanced differentiation

Tributyrin

- Highly bioavailable form of butyrate
- Superior half-life and increase in plasma levels compared to supplementation with sodium butyrate.
- Sustained increase in plasma levels of butyrate with tributyrin supplementation may allow for more systemic and epigenetic effects of butyrate.
- Multiple models suggest tributyrin protects the liver and intestinal barrier from a variety of insults, including ethanol and bacterial toxins.





Trybutyrin Complex

- Innovative trademarked ingredient
- Direct butyrate generator
- Able to produce high levels of butyrate directly in the colon.

In pre-clinical studies of Butyragen™ Tributyrin Complex has been shown to:

- Generate butyrate and other short chain fatty acids
- Significantly lower gas production when compared to other prebiotics
- Help reduce oxidative stress

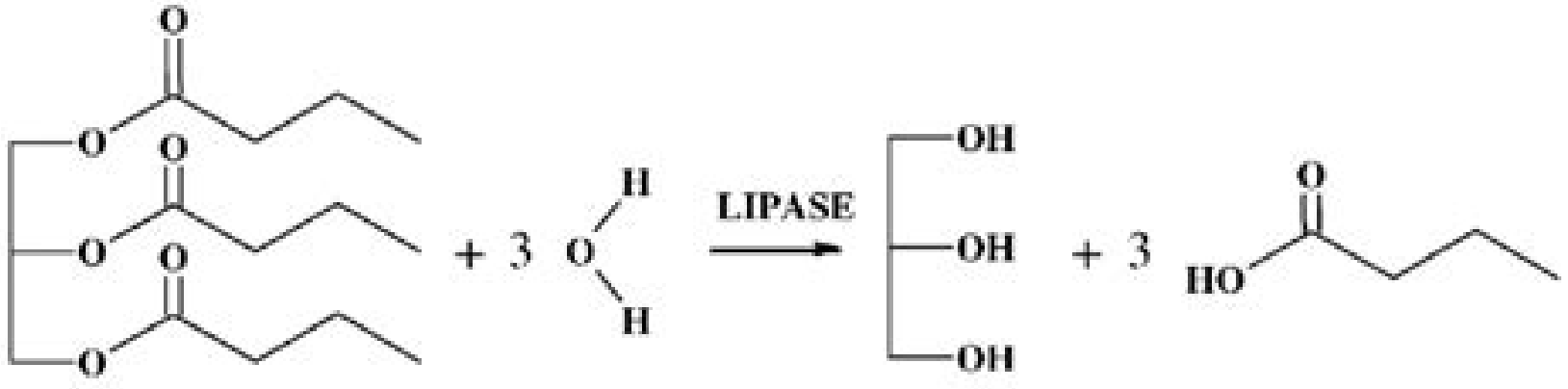
Prebiotic AND Postbiotic

- **Prebiotic** - a substrate that is selectively utilized by host microorganisms conferring a health benefit (2016)

- **Postbiotic** – preparation of inanimate microorganisms and/or their components that confers a health benefit on the host (2019)

Tributyrin Complex

- Tributyrin - three butyrate moieties esterified with glycerol
- Rapidly absorbed and hydrolyzed to butyrate by pancreatic lipases.
- Found in honey and dairy products such as butter
- Considered a “prodrug” of butyrate, with several advantages when compared to supplementation with butyrate.



Tributyrin Glycerol Butyric Acid

Tributyrin Complex

- More bioavailable
  - It is not clear that oral butyrate reaches colonocytes, as most of it is taken up by enterocytes in the proximal intestine.
- Better tolerated
- Significantly higher plasma levels
- Approximately three times the serum half-life (demonstrated in animal studies).

Tributyrin Complex

- Overcomes the limitations imposed by the short half-life of butyrate
- Promising results for wide-ranging effects in experimental models.
- Prevents fat accumulation and liver injury following alcohol toxicity
- Tributyrin also protects the liver from damage in response to lipopolysaccharide (LPS)-induced liver injury
- Down-regulates NF-κB, a critical regulator of inflammation
- Prevents oxidative stress in the colon and preserve intestinal immune function in response to ethanol-induced injury
- Modulates of the Gut-Brain Axis through PPARγ and AMPK (also supports epithelial tight junctions)
- Protects from toxins, such as C. difficile toxins, also appears to be another mechanism by which butyrate reduces intestinal inflammation

Int J Cancer. 2000 Oct 15;88(2):245-51.

Tributyrin induces differentiation, growth arrest and apoptosis in androgen-sensitive and androgen-resistant human prostate cancer cell lines

Tributyrin – Beyond the Gut

Our results demonstrate that tributyrin is more potent than butyrate in regard to cell growth inhibition and apoptosis induction at pharmacologically relevant concentrations. Hence, tributyrin may be a promising candidate for clinical protocols in prostate cancer.

The Obvious Choice

Supplement Facts

Serving Size: 2 Capsules
Servings Per Container: 30

	Amount Per Serving	% Daily Value
Vitamin A (as natural mixed carotenoids and acetate)	975 mcg RAE	108%
Pantothenic Acid (as calcium pantothenate)	16.7 mg	334%
Calcium (as calcium citrate)	40 mg	3%
Magnesium (as magnesium citrate)	40 mg	10%
ButyraGen™ Tributyrin Complex	1,000 mg	*

***Daily Value not established**

Other ingredients: Capsule shell (gelatin and water), gum arabic, guar fiber, rosemary extract, silica and cellulose.

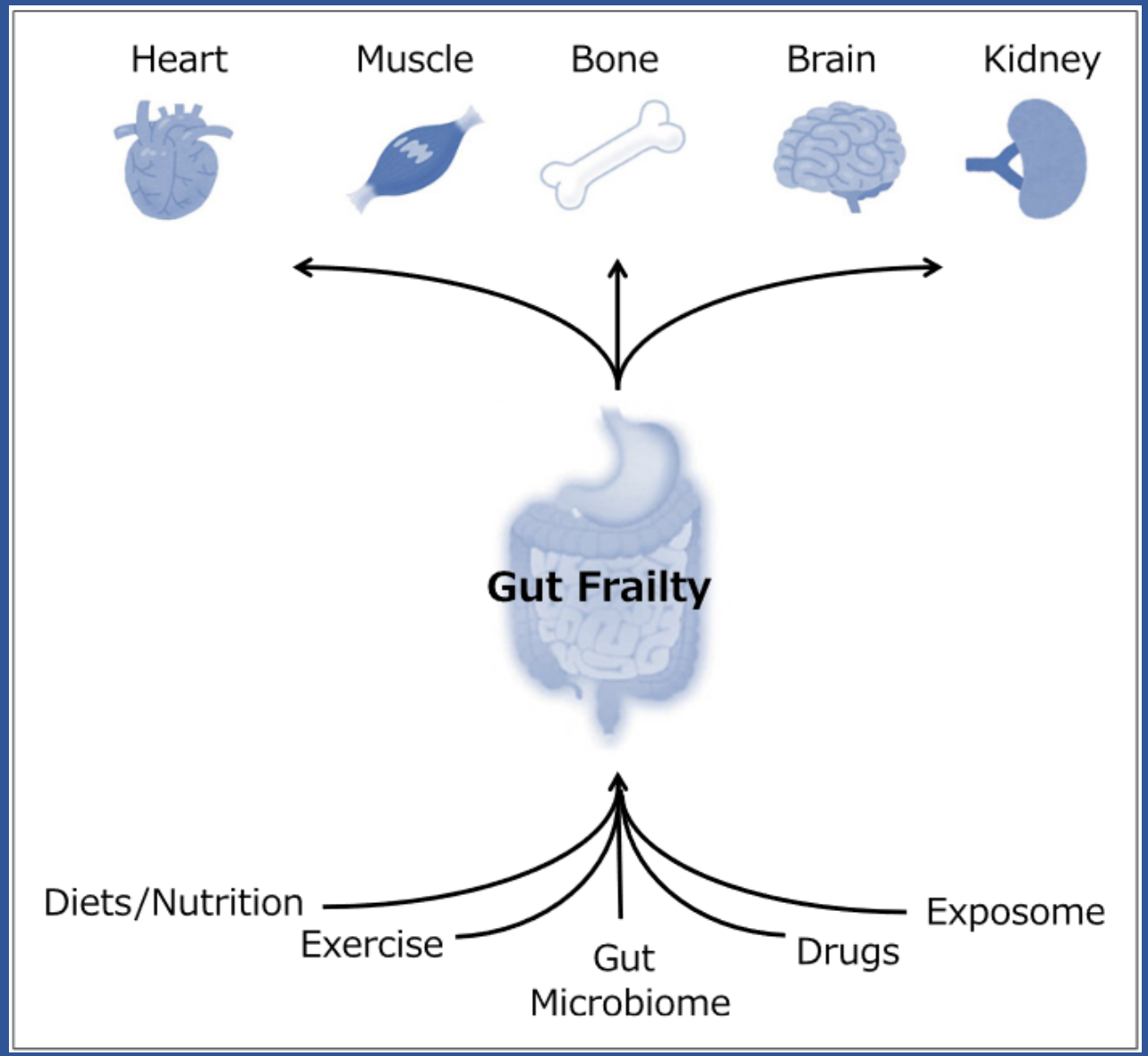
This product is gluten and dairy free.

ButyraGen™ is a trademark of NutriScience Innovations LLC.

RECOMMENDATION: Two (2) capsules as a dietary supplement or as otherwise directed by a healthcare professional.

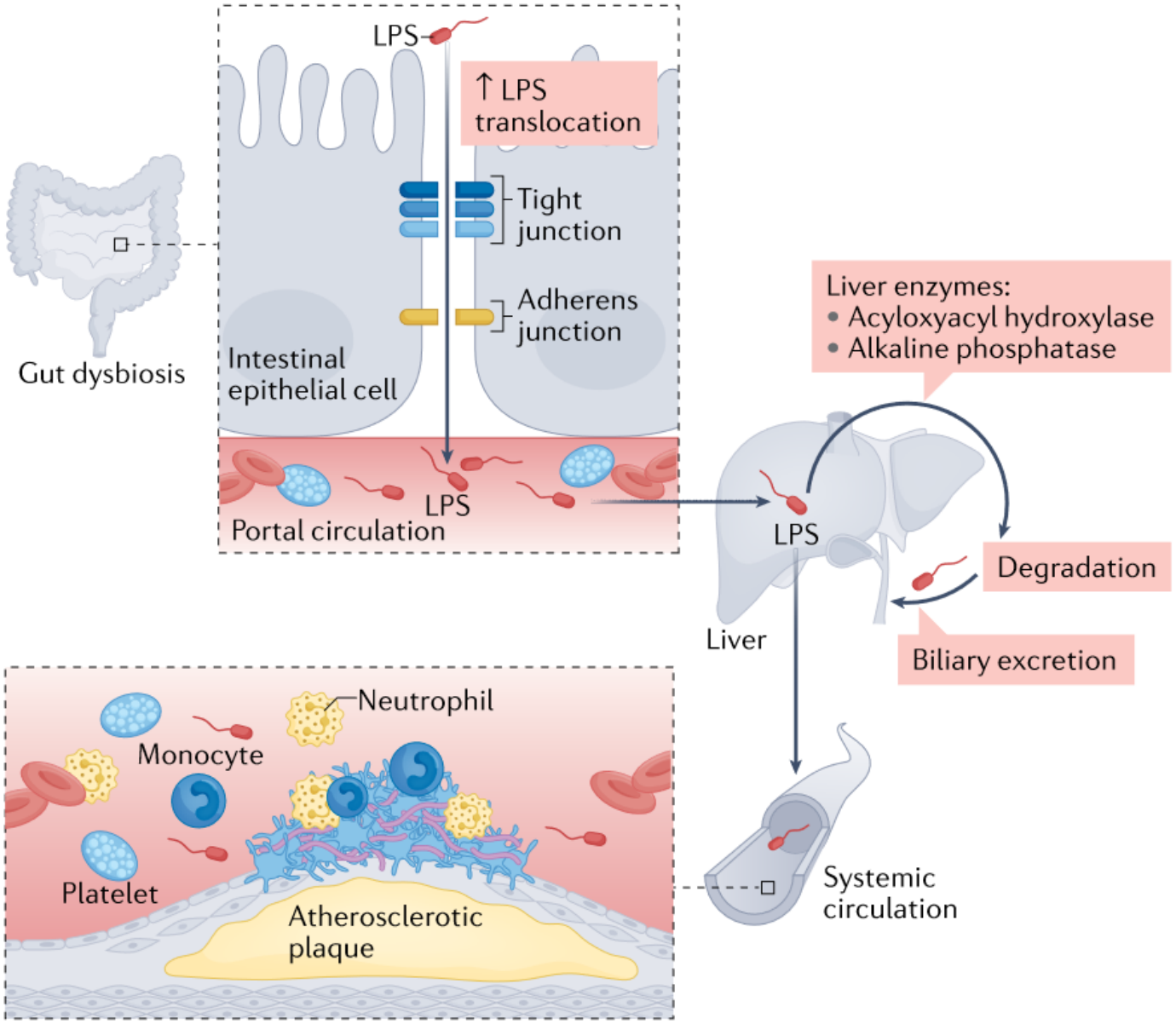
KEEP OUT OF REACH OF CHILDREN
Store in a cool, dry area. Sealed with an imprinted safety seal for your protection.

Product # 7810 Rev. 01/23


Heart
Muscle
Bone
Brain
Kidney
Gut Frailty
Diets/Nutrition
Exercise
Gut
Microbiome
Drugs
Exposome

Endotoxemia

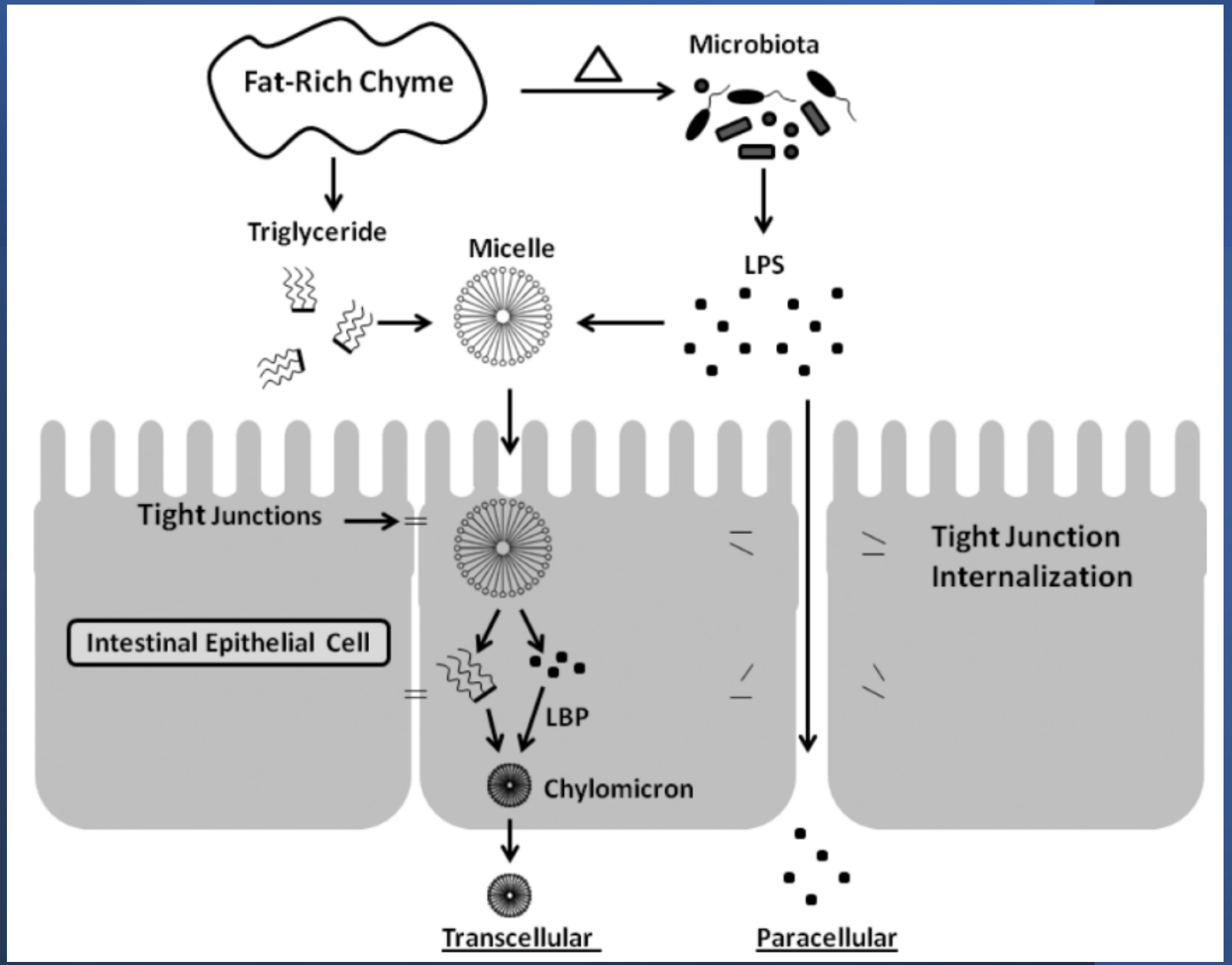
- Secondary to gut dysbiosis
- Lipopolysaccharide (LPS) translocation into the systemic circulation
  - LPS = super-antigen
- Develop of low grade endotoxemia
- Induces systemic inflammatory state



Food for Endotoxemia

- Meals influences the uptake of endotoxins to the blood
- Fat promotes uptake of endotoxins
- Dietary fiber restricts uptake
- LPS presents in the chylomicrons carrying dietary fat
  - Lymphatic uptake route
  - LPS acts like dietary fat
  - Uptake occurs over 3-4 hours – delayed reactivity
- Intestinal absorption and systemic distribution
  - Transcellular uptake through the enterocytes
  - Occurs within 30 minutes – immediately reactive
  - Possible paracellular pathway mediate through tight junction activity

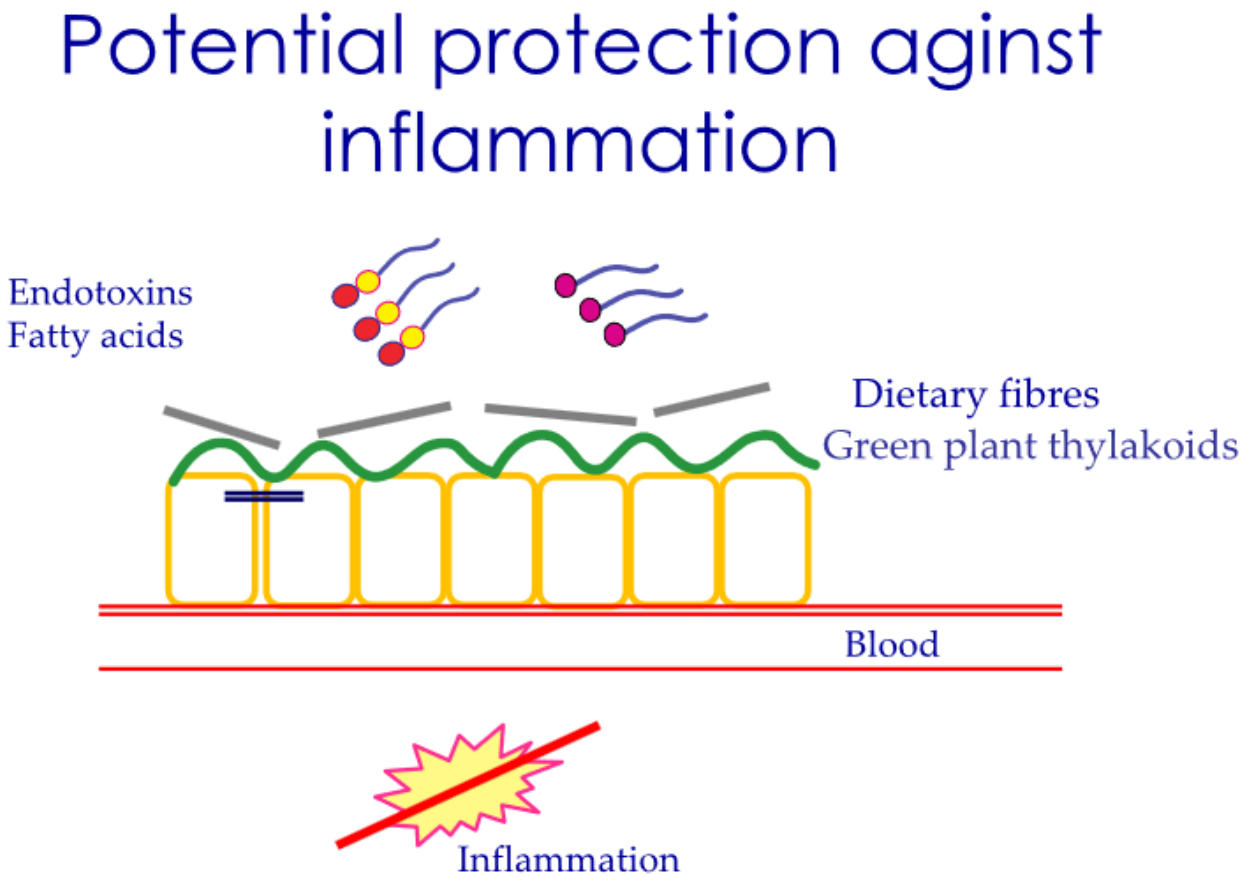
Int. J. Mol. Sci. 2021, 22, 9562



Nutr Clin Pract. 2012 April ; 27(2): 215–225.

Nutrients That Protect LPS Uptake

- Dietary fiber – delays fat digestion and absorption
- Thylakoids – extracted from green leaves
  - Delay fat digestion
  - Form a layer on the intestinal mucosa
- Restrict uptake of LPS



Problems with this Data

- Most studies combine high carbohydrate intake with fat intake
  - Often grains
- Accompanying intake of refined foods
  - Margarine
- Not all fats are created equal
  - Long chain vs. medium vs. short
  - Carbon length could make a difference
    - 16 carbon – palmitic acid
    - 18 carbon – stearic acid
  - Dietary saturated fats are not 100% saturated fat

Caveat Supported

- Evidence suggested that a fat-rich diet, depending on its quality, quantity and concomitant healthy food components, could influence metabolic endotoxemia.

- Nutrients 2019, 11, 1887

Gut-Derived Endotoxemia and CVD

- 5 year follow up
  - LPS >50 pg/ml – 3x risk of incident carotid artery atherosclerosis co
- 10 year follow up
  - LPS – binding protein significantly associated with incident CVD
- Association between circulating LPS levels and myocardial infarction
- Coronary ischemia is associated with intestinal barrier dysfunction
- LPS are a trigger for atrial fibrillation
- Circulating LPS level lab marker associations
  - Triglycerides, total cholesterol, fasting glucose, insulin, HbA1c and CRP

Nat Rev Cardiol. 2023 Jan;20(1):24-37.

LPS Circulation

LPS in circulation is mostly bound to lipoproteins (80–97%)

- Highest concentration in LDL (35.7%)
- Lowest in VLDL (13.9%)

LPS enters arterial wall bound to pro-atherogenic lipoproteins

- Favors LDL oxidation
- Propagation of arterial inflammation

LPS destabilizes atherosclerotic lesions

- Plaques become more vulnerable to rupture and erosion
- Activation in atherosclerotic lesions of the arachidonic acid pathway that leads to biosynthesis of leukotrienes

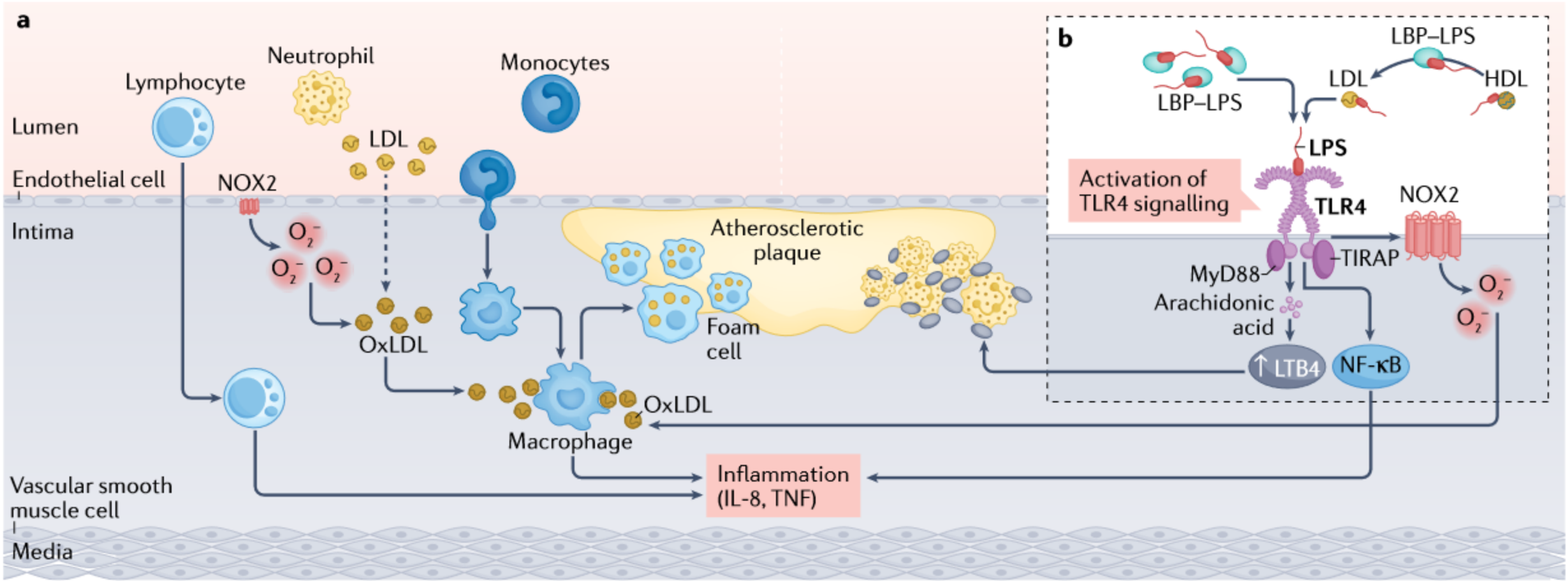


Fig. 3 | **Mechanisms of LPS-mediated atherosclerosis. a** | LDL can cross into the arterial wall and undergo oxidation in the subendothelial space, leading to the formation of oxidized LDL (oxLDL). OxLDL is taken up by macrophages, inducing foam cell formation and inflammatory cytokine production. **b** | Lipopolysaccharides (LPS) can cross into the arterial wall, either together with LPS binding protein (LBP) or by LBP-mediated LPS transfer from HDL to LDL particles. LPS binds to Toll-like receptor 4 (TLR4) in several cell types, leading to phosphorylation of Toll–interleukin-1 receptor domain-containing adaptor protein (TIRAP) and recruitment of the myeloid differentiation primary response protein 88 (MyD88) to the cytoplasmic domain of TLR4. Downstream signalling induces the activation of the transcription factor nuclear factor-κB (NF-κB), which increases the production of pro-inflammatory cytokines, such as IL-8 and tumour necrosis factor (TNF); oxidative stress via upregulation of NADPH oxidase 2 (NOX2)-derived reactive oxygen species, which further promotes LDL oxidation; and destabilization of the atherosclerotic plaque via activation of the arachidonic acid pathway and biosynthesis of leukotriene B_4 (LTB4), which attract leukocytes to the atherosclerotic lesion. O_2^-, superoxide.

CVD-CKD-LPS Link

- Pathway to chronic kidney disease (CKD)
- Hypertension
  - Important risk factor for CKD
  - Leads to high intraglomerular pressure
  - 85-90% patients with stage 3-5 CKD
- CKD
  - No cure
  - Poor and unrecognized prevention strategies
  - Lack of symptoms in early stage
  - Intervention options – dialysis and kidney transplant

Nat Rev Nephrol. 2018 July ; 14(7): 442–456.

Vagal Nerve Communication

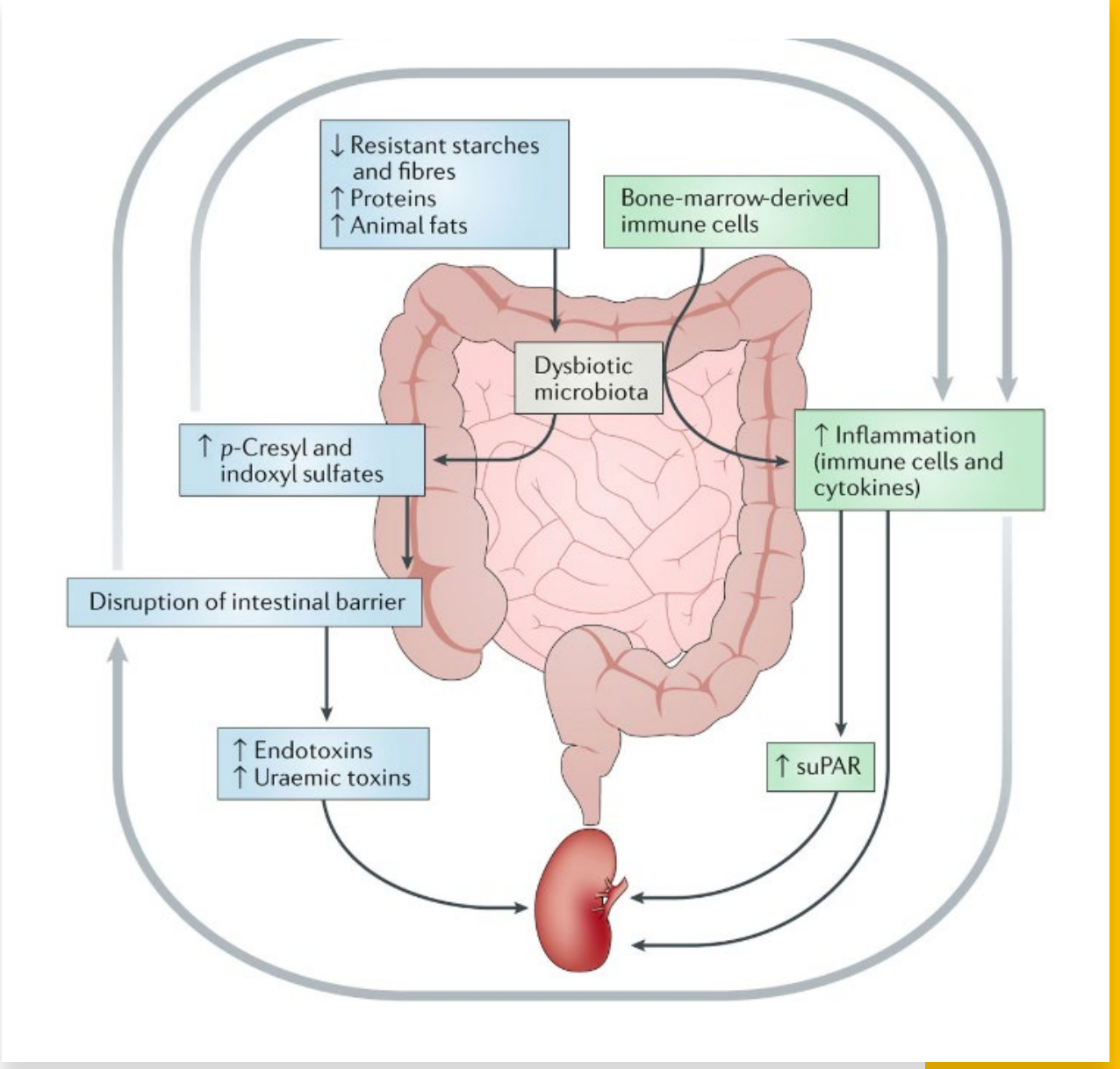
Facilitates bidirectional communication between the brain and enteric nervous system

Expresses receptors that sense SCFA

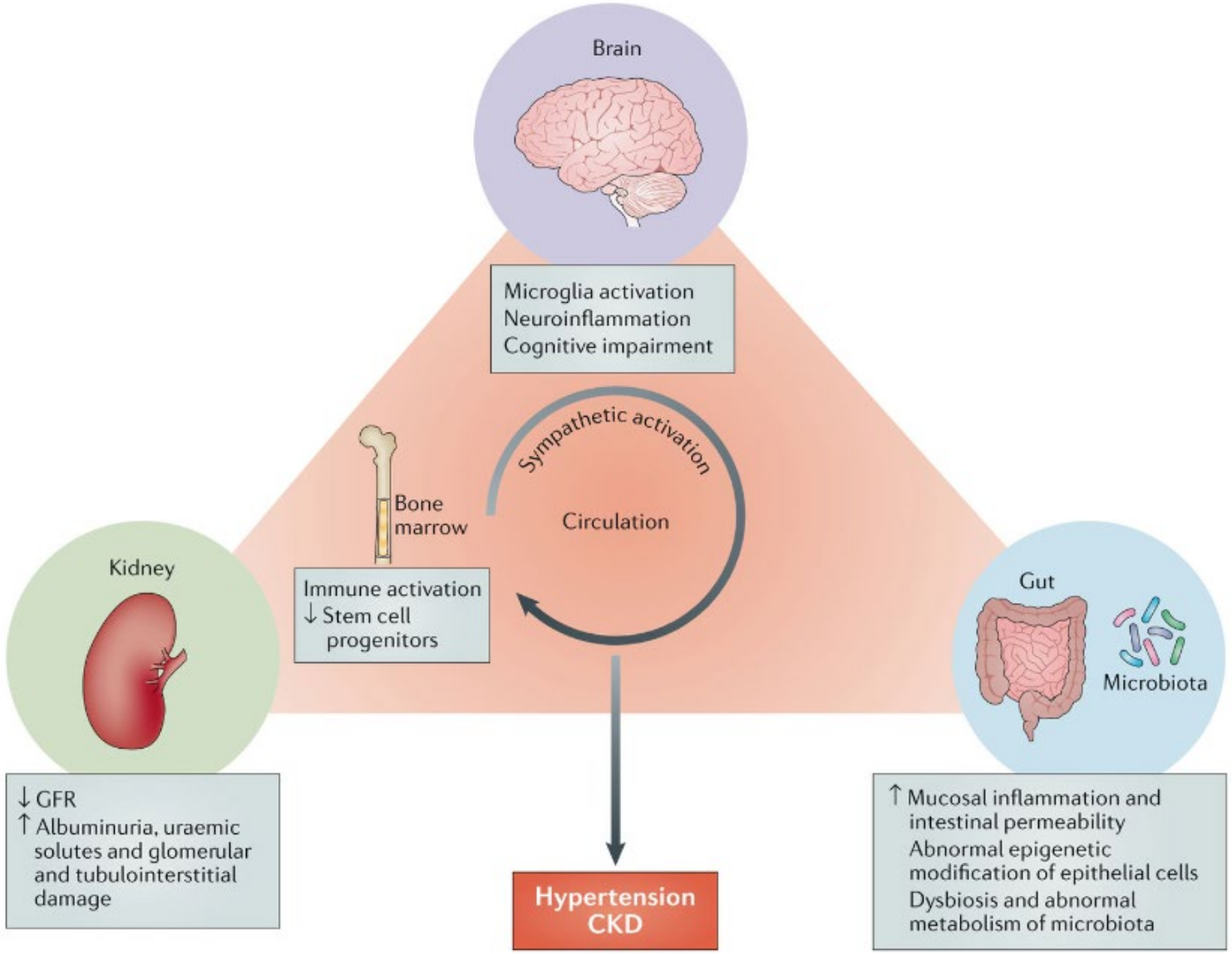
Modulated by Lactobacillus rhamnosus and Bifidobacter longum

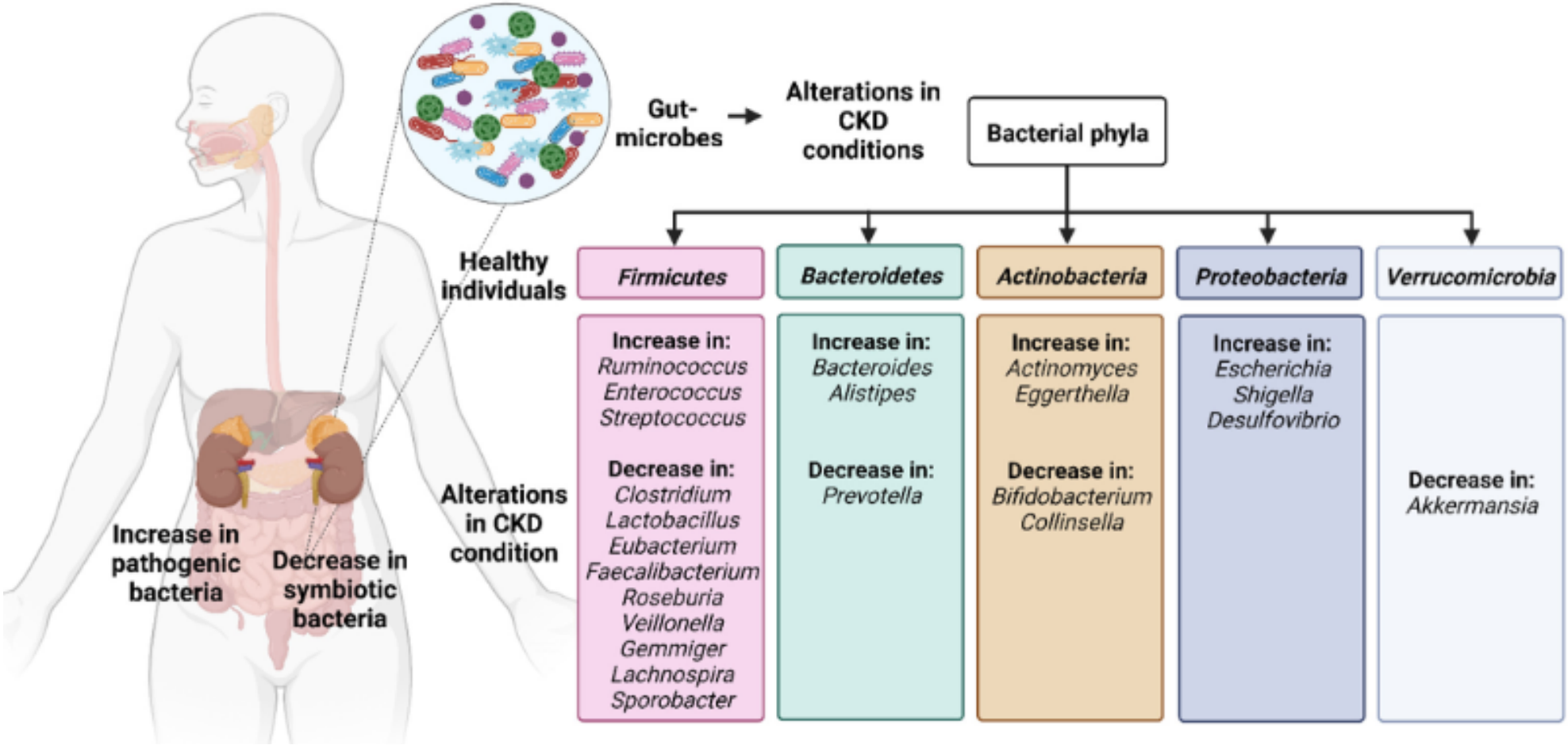
Gut Microbiota in Hypertension

- Dysregulation causes hypertension
  - Renin-angiotensin system
  - Autonomic nervous system
  - Immune system
  - Gut dysbiosis
    - Induces immune and autonomic changes
    - Gut-kidney axis
      - Metabolism dependent and immune pathways



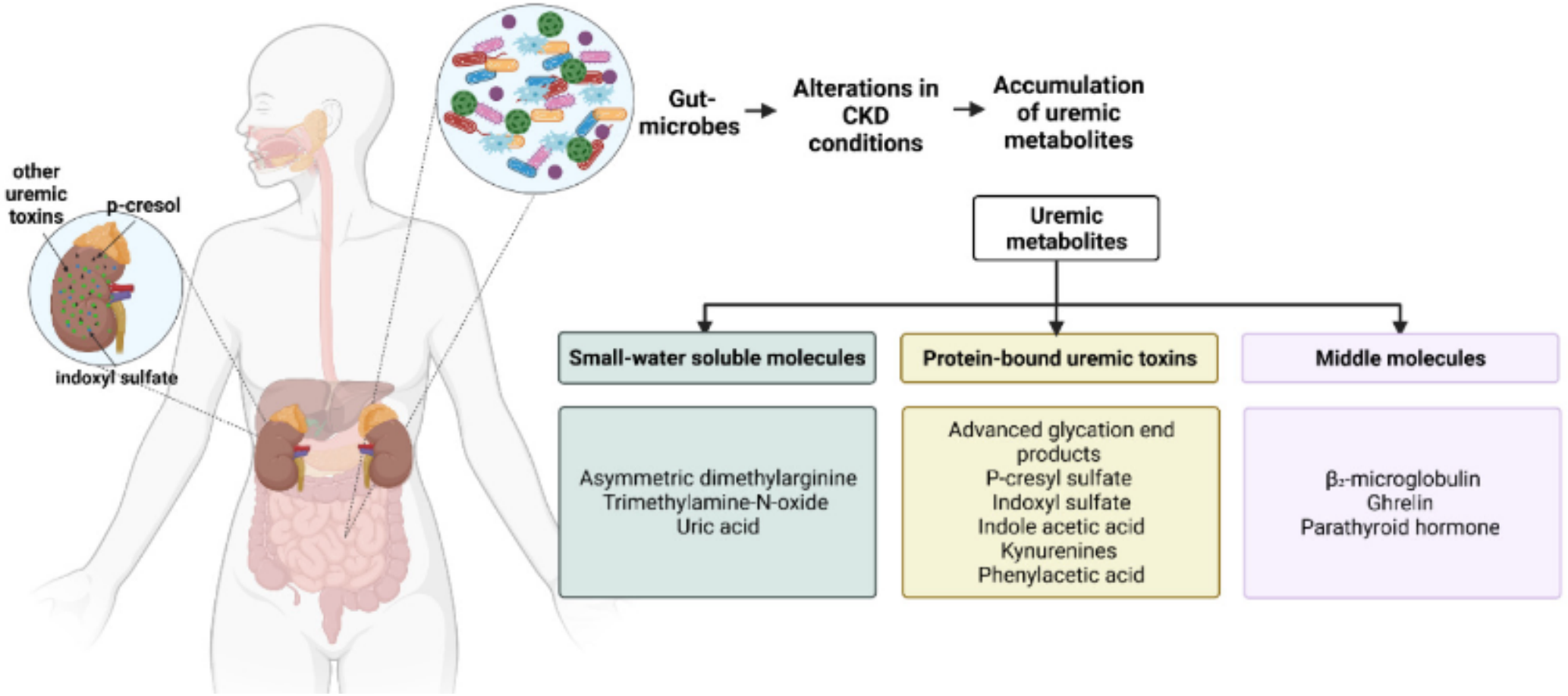
Brain-Gut-Kidney Axis





Microbial Profile Breakdown in CKD

Measurable Uremic Metabolites to Track



Small Water-Soluble Molecules

Asymetric Dimethyl Arginine (ADMA)

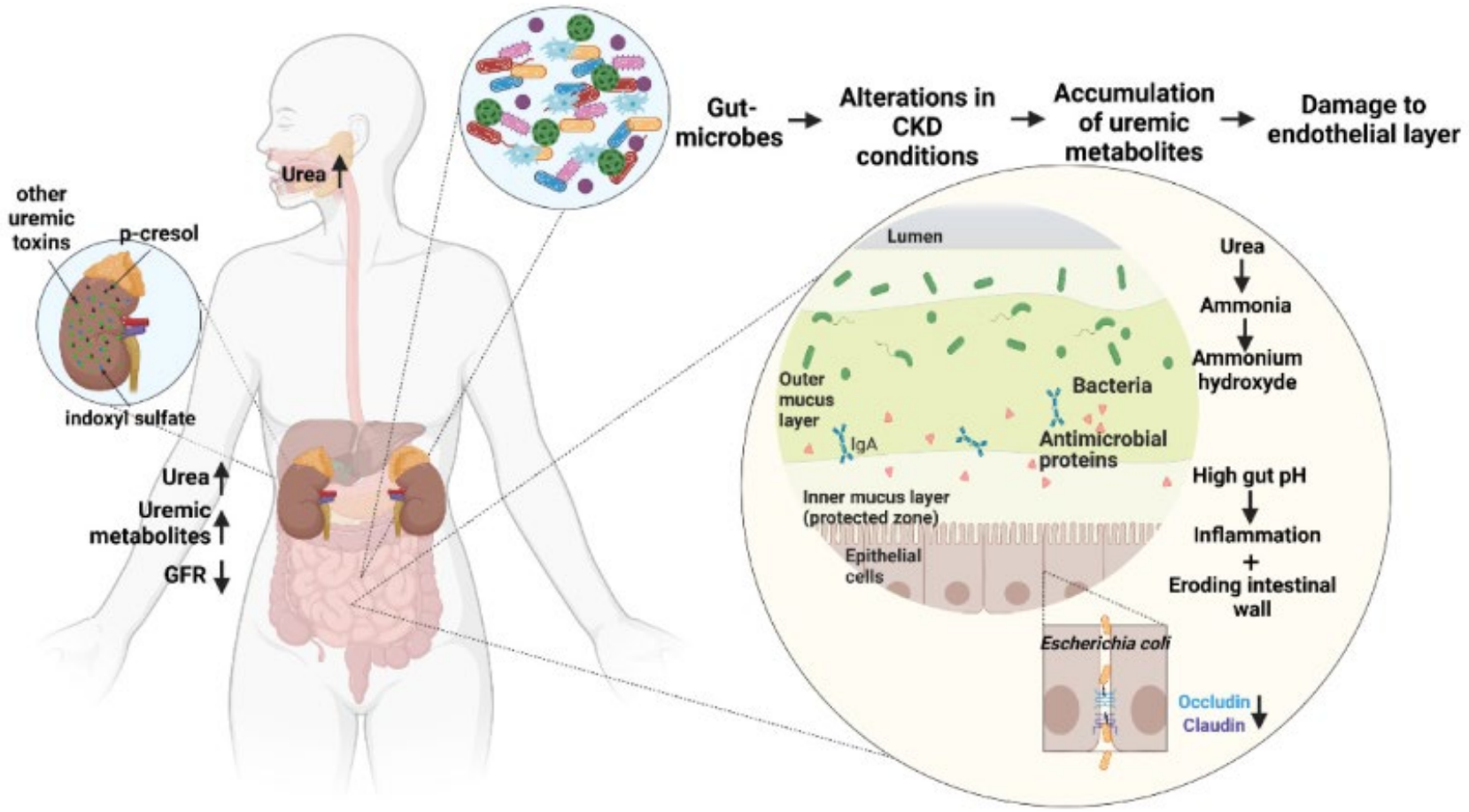
- Analogue of arginine
- Downregulates NO production
  - Increased vaso-contrictive tone
  - Impacts microcirculation of all major organs
- Independent risk factor for all cause mortality

Trimethylamine-N-oxide (TMAO)

- Questionable association

Urea

- Relevant gut-derived toxin
- Can trigger insulin resistance
- Endothelial dysfunction via free radical generation



Urea Leads to Further Intestinal Damage

Gut as a Therapeutic Target for CKD

toxins

MDPI

Review

Homeostasis in the Gut Microbiota in Chronic Kidney Disease

Shruti Bhargava [1], Erik Merckelbach [1], Heidi Noels [1], Ashima Vohra [2] and Joachim Jankowski [1,3,*]

toxins

MDPI

Review

Microbiome in Chronic Kidney Disease (CKD): An Omics Perspective

Sonnal Lohia [1,2], Antonia Vlahou [1] and Jerome Zoidakis [1,*]

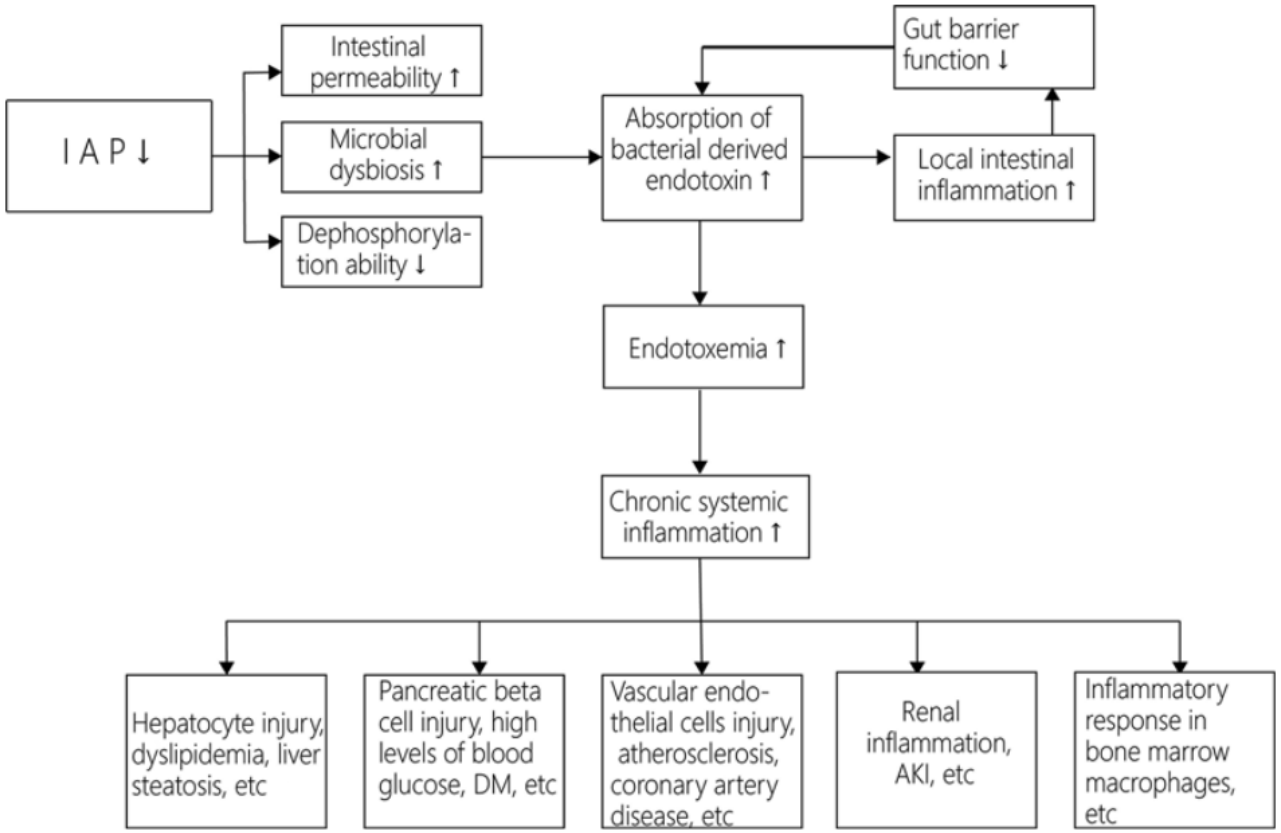
Nat Rev Nephrol. 2018 July ; 14(7): 442–456. doi:10.1038/s41581-018-0018-2.

The gut microbiota and the brain-gut-kidney axis in hypertension and chronic kidney disease

Tao Yang[1], Elaine M. Richards[1], Carl J. Pepine[2], and Mohan K. Raizada[1,*]

Endotoxemia –Intestinal Alkaline Phosphatase

- Aging – gut barrier dysfunction and gut-derived chronic inflammation
- Alkaline phosphatase
  - Multiple Isoenzymes (bone, liver, gut)
  - Gut brush border enzyme
  - Inhibits inflammatory mediators like LPS
  - Supports gut barrier function
  - Regulates microbial homeostasis



Intestinal Alkaline Phosphatase (IAP)

- Dephosphorylates LPS, flagellin, and DNA
- Reduces LPS toxicity by 100-fold
- Supports bicarbonate secretion in the duodenum to increase pH and reduces acidity as part of negative feedback loop
- Though mostly membrane bound, a small amount of IAP is bidirectionally released into the blood as well as into the lumen
- Serum alkaline phosphatase is a reasonable proxy
- Zinc dependent

IAP and Gut Axes

	IAP deficiency indicates and/ or causes these dysfunctions	IAP supplementation prevents and/or attenuate these dysfunctions
Gut	NEC [31] and IBD [13]	*Salmonella* and *C. difficile* infection [24, 26], NEC [29, 30], and UC [32]
Gut-liver axis	Aging-related liver change [10] and cirrhosis [34]	ALD [36] and cirrhosis [34]
Gut-pancreas axis	T2DM [48]	Metabolic syndrome [10]
Gut-heart axis	IHD [50]	AMI-induced inflammation [55], myocardial dysfunction [53], and complications of AMI [53]
Gut-kidney axis		Sepsis-induced AKI [61], renal inflammation [66], and damage of I/R-induced AKI [66]
Gut-bone axis	IAP deficiency increases cortical thickness, the volume of intracortical bone, and trabecular connectivity [70]	

AMI, myocardial infarction; ALD, alcoholic liver disease; I/R, ischemia-reperfusion.

Targeting the Intestinal Barrier to Prevent Gut-Derived Inflammation and Disease: A Role for Intestinal Alkaline Phosphatase

Florian Kühn[a] Ruifeng Duan[a] Matthias Ilmer[a] Ulrich Wirth[a]
Fatemeh Adiliaghdam[b] Tobias S. Schiergens[a] Joachim Andrassy[a]
Alexandr V. Bazhin[a] Jens Werner[a]

[a]Department of General, Visceral and Transplant Surgery, University Hospital of LMU Munich, Munich, Germany;
[b]Department of Surgery, Massachusetts General Hospital, Harvard Medical School, Boston, MA, USA

Keywords
Intestinal alkaline phosphatase · Lipopolysaccharides · Gut barrier · Endotoxemia · Microbiome · Diabetes · Metabolic syndrome

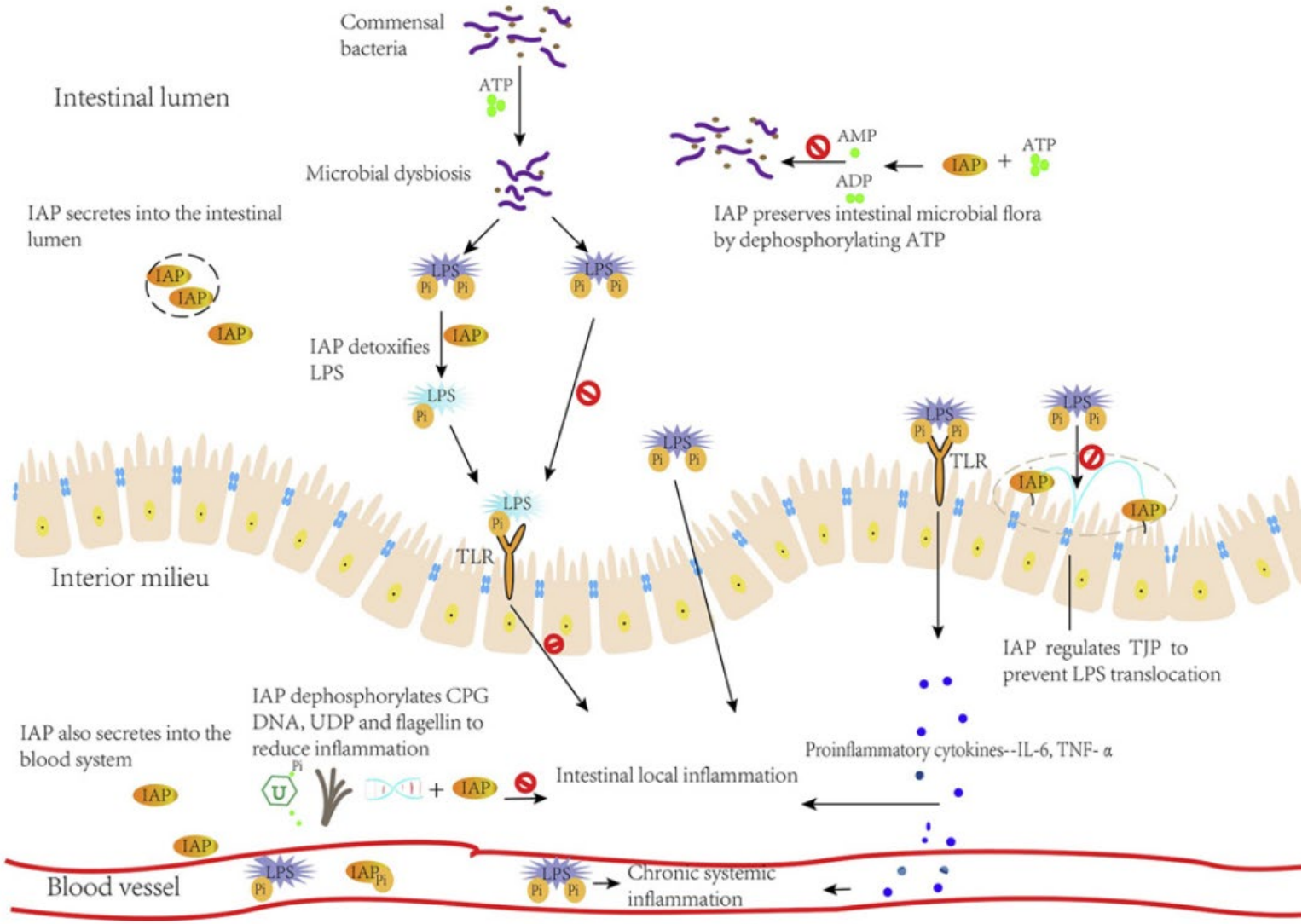
Abstract

Background: Intestinal alkaline phosphatase (IAP) as a tissue-specific isozyme of alkaline phosphatases is predominantly produced by enterocytes in the proximal small intestine. In recent years, an increasing number of pathologies have been identified to be associated with an IAP deficiency, making it very worthwhile to review the various roles, biological functions, and potential therapeutic aspects of IAP. ***Summary:*** IAP primarily originates and acts in the intestinal tract but affects other organs through specific biological axes related to its fundamental roles such as promoting gut barrier function, dephosphorylation/detoxification of lipopolysaccharides (LPS), and regulation of gut microbiota. ***Key Messages:*** Numerous studies reporting on the different roles and the potential therapeutic value of IAP across species have been published during the last decade. While IAP deficiency is linked to varying degrees of physiological dysfunctions across multiple organ systems, the supplementation of IAP has been proven to be beneficial in several translational and clinical studies. The increasing evidence of the salutary functions of IAP underlines the significance of the naturally occurring brush border enzyme.

Introduction

As an isoenzyme of alkaline phosphatase (AP), intestinal AP (IAP) is predominantly produced and secreted by enterocytes in the duodenum, and at lower levels in the jejunum, ileum, and colon [1]. IAP molecules can be found in high concentrations within luminal vesicles secreted by enterocytes on the brush border of the microvilli. Though mostly membrane bound, a small amount of IAP is bidirectionally released into the blood as well as into the lumen [2]. IAP functions to inhibit lipopolysaccharides (LPS) and other inflammatory mediators responsible for endotoxemia and appears to be an important positive regulator of gut barrier function and microbial homeostasis (Fig. 1). Based on the functional roles for IAP, in particular its salutary role in gut barrier integrity and its ability to inhibit bacteria-derived inflammatory mediators, its deficiency has been linked to several pathologies and disorders (Fig. 2). The functions of IAP have been unraveled in several basic, translational, and clinical studies underlining the importance of this gut brush border enzyme. The purpose of this review is to discuss the various roles of IAP within the gut and its connected axes.

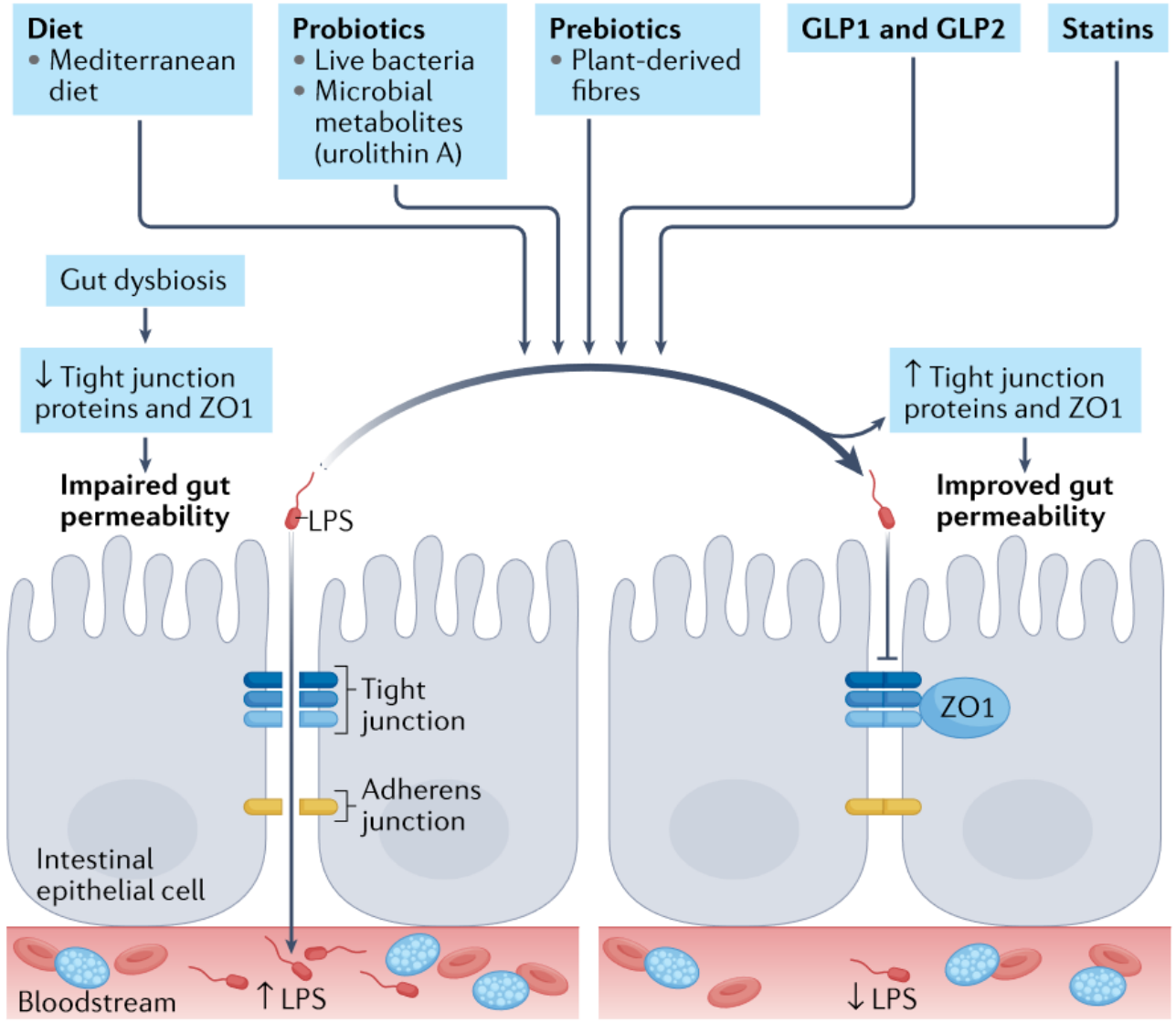
F.K. and R.D. share first authorship.



Visc Med 2021;37:383–393

Therapeutic Considerations

- Mediterranean Diet
- SCFA
- Prebiotic fiber
- Polyphenols
- Omega-3s
- Probiotics
  - Lactobacillus plantarum
  - Lactobacillus rhamnosus
  - Akkermansia mucinophilia
- Vitamin D



Decreasing LPS

Glutamine

- In the study by Abboud et al., obese or overweight subjects ingested 30 g of glutamine per day for eight weeks (*Nutrients 2019, 11, 536*.)
- Blood LPS levels and waist circumference decreased

Prebiotics

- Oligofructose (glucose, fructose) – fruits and vegetables
- Inulin – fruits and vegetables
- Galacto-oligosaccharides (glucose, galactose) – dairy

Sulfated Polysaccharides

- Chondroitin sulfate

Inflammaging and Sarcopenia

Sarcopenia and Inflammaging

- Sarcopenia - progressive and generalized loss of skeletal muscle mass and function
- Muscle weakness (dynapenia)
- Inflammaging as a cause
  - Direct and indirect drivers
    - Cellular senescence, immunosenescence, alterations in adipose tissue, damage-associated molecular patterns (DAMPs), and gut microbes
- Sarcopenia causes inflammaging
- Vicious cycle

Sarcopenia Benchmarks

- Muscle strength
  - Hand grip strength
    - Male - <28kg
    - Female - <18kg
- Physical performance
  - 6 meter gait speed <1.0 m/sec
  - 5 time chair stand test ≥ 12 sec
  - Short physical performance test ≤ 9 sec
- Appendicular skeletal muscle mass
  - DXA – ($M<7.0kg/m^2$, $F<5.4kg/m^2$)
  - BIA - ($M<7.0kg/m^2$, $F<5.7kg/m^2$)

J Korean Med Sci. 2022 May 9;37(18):e146.

Diagnostic Criteria by Group

- EWGSOP, European Working Group on Sarcopenia in Older People
- IWGS, International Working Group on Sarcopenia
- AWGS, Asian Working Group for Sarcopenia
- FNIH, Foundation for the National Institutes of Health
- SSCWD, Society on Sarcopenia, Cachexia and Wasting Disorders
- SMI, Skeletal Muscle Mass Index
- BMI, Body Mass Index
- DXA, Dual Energy X.

Diagnostic Criteria	Skeletal Muscle Mass index	Grip Strength (Kg)	Gait Speed (m/s)
EWGSOP 2010 (4)	SMI (Kg/m^2) DXA male<7.0, famle<6.0	male<27, famle<16	≤0.8 (4m)
EWGSOP 2018 (5)	SMI (Kg/m^2) DXA male<7.0, famle<6.0	male<27, famle<16	≤0.8 (4m)
IWGS (8)	SMI (Kg/m^2) DXA male<7.23, famle<5.67	–	<1.0
AWGS2014 (9)	SMI (Kg/m^2) DXA male<7.0, famle<5.4 BIA male<7.0, famle<5.7	male<20, famle<15	<1.0 (6m)
AWGS2019 (10)	SMI (Kg/m^2) DXA male<7.0, famle<5.4 BIA male<7.0, famle<5.7	male<20, famle<15	<1.0 (6m)
FNIH (11)	DXA Appendicular Skeletal Muscle Mass/BMI (Kg/BMI) male<0.789, famle<0.512	male<26, famle<16	≤0.8
SSCWD (12)	Average level -2 standard deviations of healthy individuals aged 20-30 from the same ethnicity.	–	<1.0 or <400m (6min)

Risk Factors

Lack of exercise – foremost risk factor

- Gradual decline in muscle fiber numbers begins around age 50
- More pronounced with inactivity

Decreases in anabolic hormones

- Testosterone, growth hormone, thyroid hormone, insulin-like growth factor, vitamin D

Increases in catabolic signals

- Pro-inflammatory cytokines
- Tumor necrosis factor alpha (TNFα) and interleukin-6 (IL-6)

Protein synthesis

- Decreased ability
- Inadequate intake of calories / protein
- Oxidized proteins leading to buildup of lipofuscin

Risk Factors - Continued

Motor unit remodeling

- Reduction in motor nerve cells
- Failure of satellite cell activation
  - Satellite cells – maintain muscle function

Genetic influence

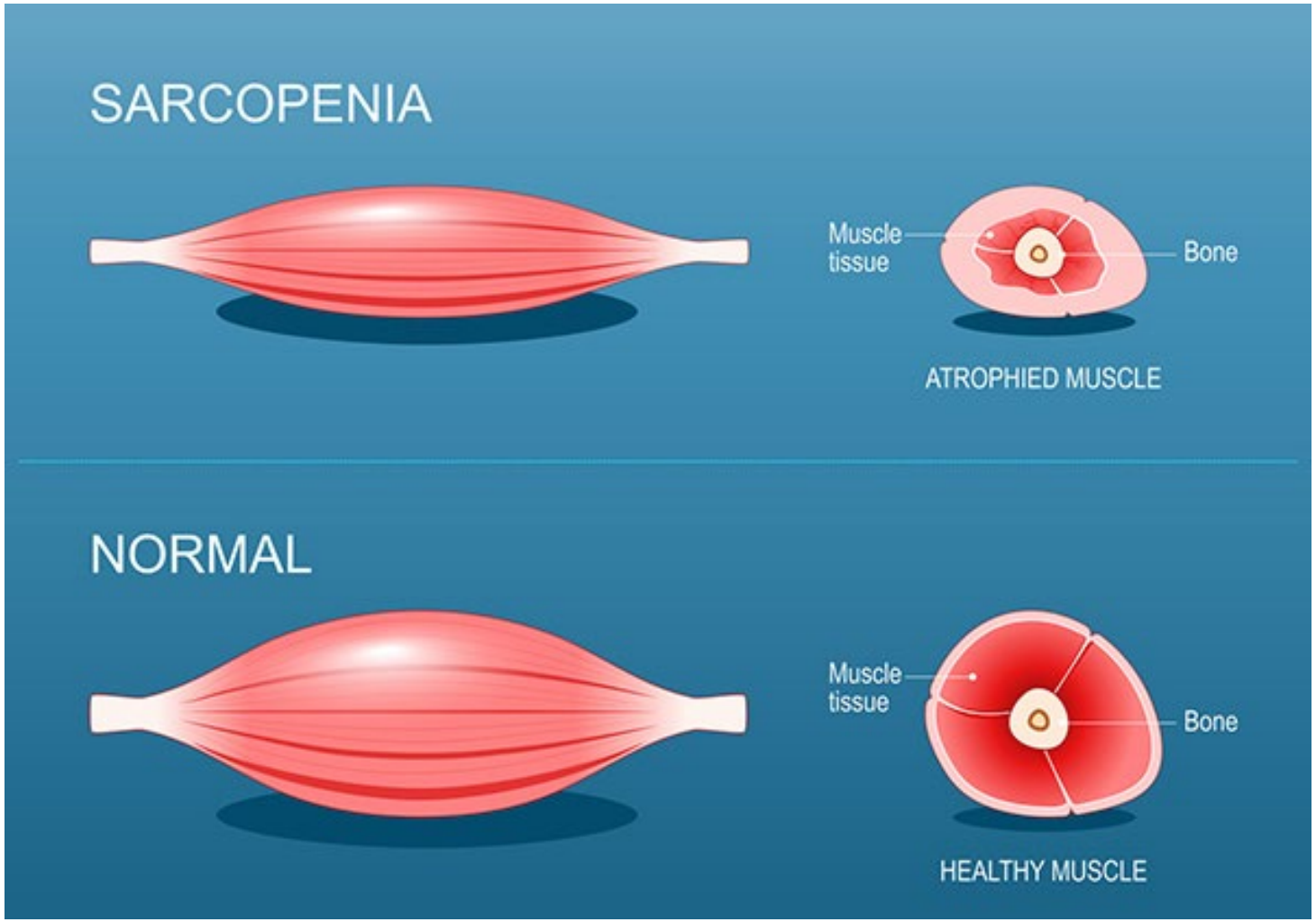
- Genes govern muscle mass and function maintenance

Early developmental influences

- Low birth weight

Types of Sarcopenia

- Primary sarcopenia - related to aging
- Secondary sarcopenia - independent of age in the context of chronic disease



Journal of Cachexia, Sarcopenia and Muscle 2021; 12: 1380–1392

Sarcopenia: A Rheumatic Disease?

Patients with systemic autoimmune diseases

- Systemic lupus erythematous (SLE)
- Rheumatoid arthritis (RA)
- Spondyloarthritides
- Systemic sclerosis

Pro-inflammatory state and decreased muscle use

Inactivity and pain

Loss of muscle mass and function is 2-3x more common

Rapid decline in hand grip strength

Rheum Dis Clin North Am. 2018 August ; 44(3): 393–404.

Sarcopenia and Type II Diabetes

- Unlike sarcopenia, T2DM-related sarcopenia is characterized by a reduction in type I fibers, and it differs from disuse muscle atrophy as well
- Defined as a secondary sarcopenia

frontiers | Frontiers in Endocrinology

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Type 2 diabetes mellitus related sarcopenia: a type of muscle loss distinct from sarcopenia and disuse muscle atrophy

Zhenchao Liu[1], Yunliang Guo[1]* and Chongwen Zheng[2]*

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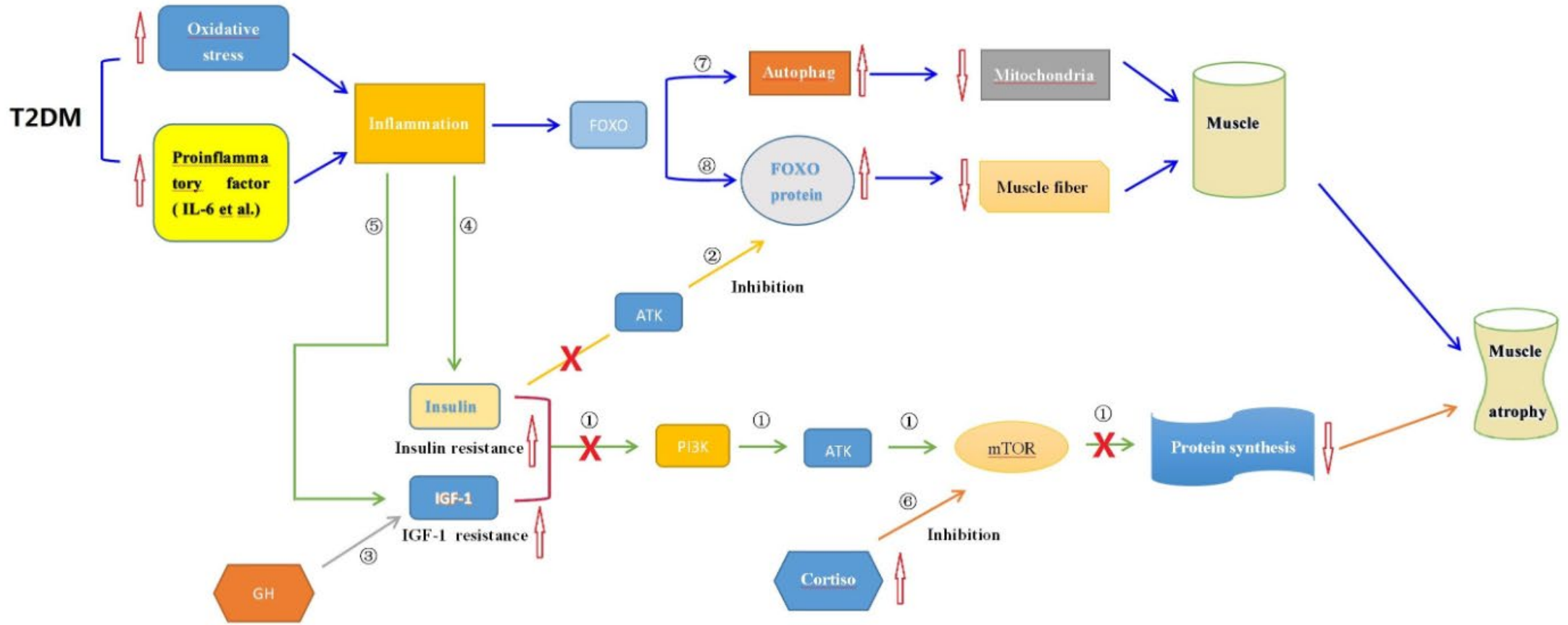
Muscle loss is a significant health concern, particularly with the increasing trend of population aging, and sarcopenia has emerged as a common pathological process of muscle loss in the elderly. Currently, there has been significant progress in the research on sarcopenia, including in-depth analysis of the mechanisms underlying sarcopenia caused by aging and the development of corresponding diagnostic criteria, forming a relatively complete system. However, as research on sarcopenia progresses, the concept of secondary sarcopenia has also been proposed. Due to the incomplete understanding of muscle loss caused by chronic diseases, there are various limitations in epidemiological, basic, and clinical research. As a result, a comprehensive concept and diagnostic system have not yet been established, which greatly hinders the prevention and treatment of the disease. This review focuses on Type 2 Diabetes Mellitus (T2DM)-related sarcopenia, comparing its similarities and differences with sarcopenia and disuse muscle atrophy. The review show significant differences between the three muscle-related issues in terms of pathological changes, epidemiology and clinical manifestations, etiology, and preventive and therapeutic strategies. Unlike sarcopenia, T2DM-related sarcopenia is characterized by a reduction in type I fibers, and it differs from disuse muscle atrophy as well. The mechanism involving insulin resistance, inflammatory status, and oxidative stress remains unclear. Therefore, future research should further explore the etiology, disease progression, and prognosis of T2DM-related sarcopenia, and develop targeted diagnostic criteria and effective preventive and therapeutic strategies to better address the muscle-related issues faced by T2DM patients and improve their quality of life and overall health.

Comparison

- Muscle Fiber Types
- Type I – Slow twitch
  - Small diameter
  - More mitochondria
  - High oxidative capacity
  - Endurance activities and posture
- Type II – Fast twitch
  - Large diameter
  - Abundant muscle proteins
  - High glycogen content
  - Explosive movements

	Sarcopenia	Disuse muscle atrophy	T2DM related sarcopenia
Primary Causes	**Aging**	**Prolonged disuse**	**T2DM**
Onset Age	Often >65 years	Any age group	Any age group
Muscle Mass	Decreased	Decreased	Decreased
Muscle Strength	Decreased	Decreased	Decreased
Muscle Fiber Area	Decreased	Decreased	Decreased
Muscle Fiber Changes	**Predominantly type II fibers**	**Predominantly type I fibers**	**Predominantly type I fibers**
Muscle Cell Count	Decreased	Unchanged	Decreased
Muscle	Difficult	Possible	Difficult

Mechanisms of Muscle Wasting Associated with T2DM



Front. Endocrinol. 15:1375610.

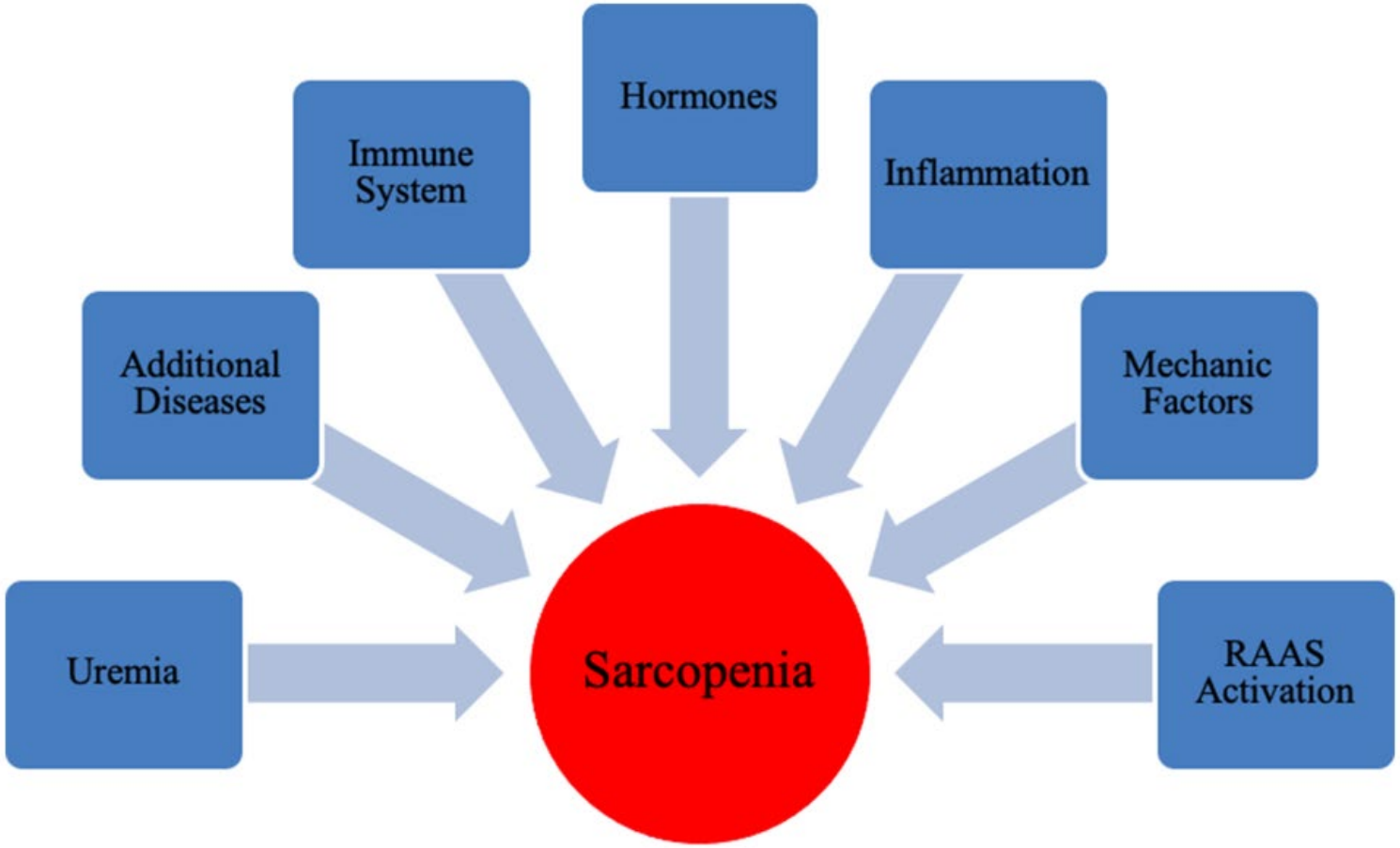
Sarcopenia and CKD

- Chronic kidney disease (CKD) is often called a model of ‘accelerated ageing’
- Frequency of sarcopenia increases as the CKD stage advances
- Loss of muscle mass in CKD may be attributed to a negative balance of protein homeostasis
  - Leads to increased catabolism and decreased synthesis of muscle
- CKD - facilitate impairments of muscle regeneration process
  - decreased production of myogenic regulatory factors and reduced cellular activation
  - Increased blood levels of myostatin (negative regulator of muscle mass)
- Impaired mitochondrial function leads to decreased muscular endurance

Sarcopenia Catabolic Activity and CKD

- Chronic inflammation
- Uremic toxins
- Malnutrition
- Hormonal imbalance
  - (insulin resistance, vitamin D deficiency, and hypogonadism)
- Oxidative stress
- Increased ubiquitination
  - Renin–angiotensin–aldosterone system is up-regulated in CKD
  - Impair muscle regeneration
  - Invoke ubiquitin proteasome system proteolytic pathways

Factors Affecting Sarcopenia Development in CKD



Uremic factors: Metabolic acidosis, dialysis-related factors, malnutrition, dietary factors.

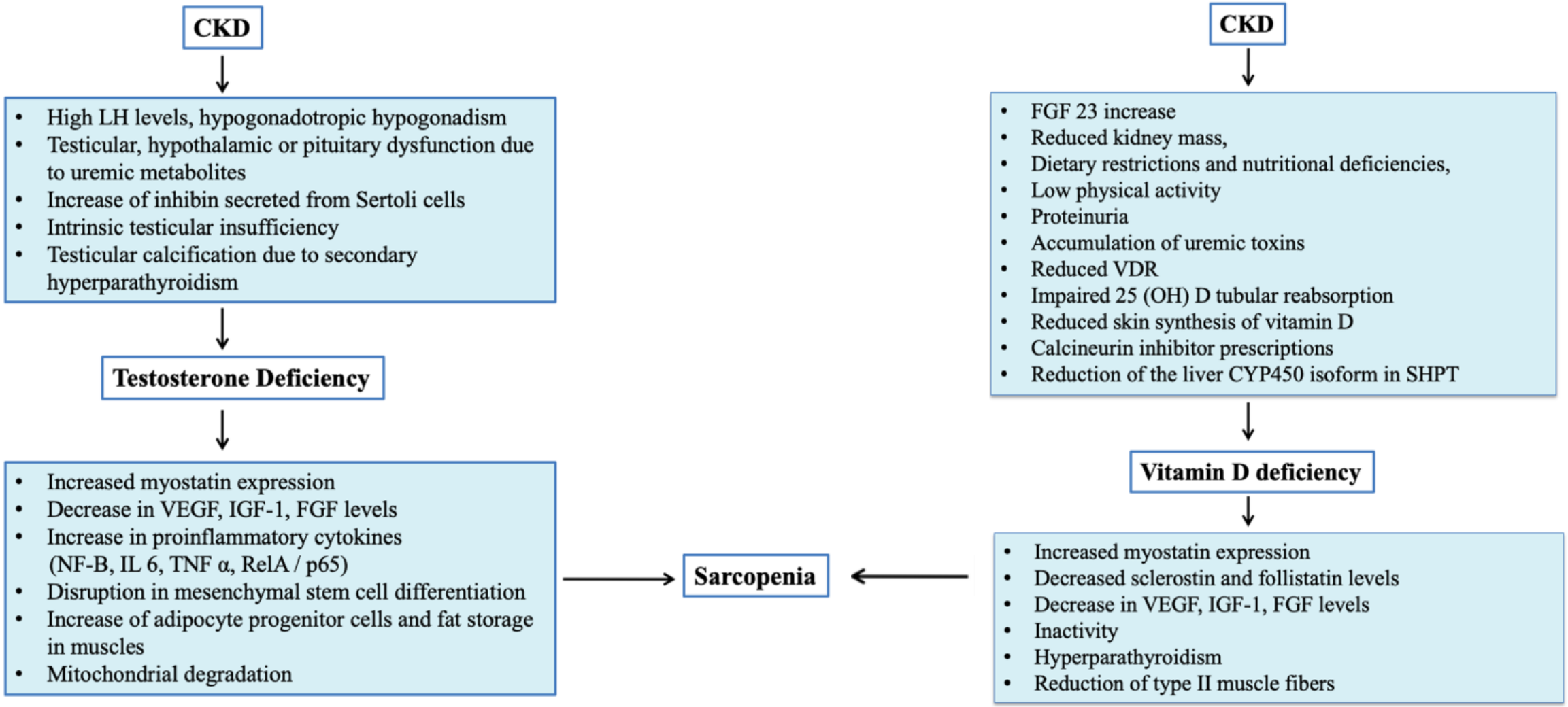
Comorbid diseases: Diabetes, cardiovascular disease, infection, malignancy

Immune system changes: Increases in inflammatory cytokines (TNF-alpha, IL-6, IL-1beta)

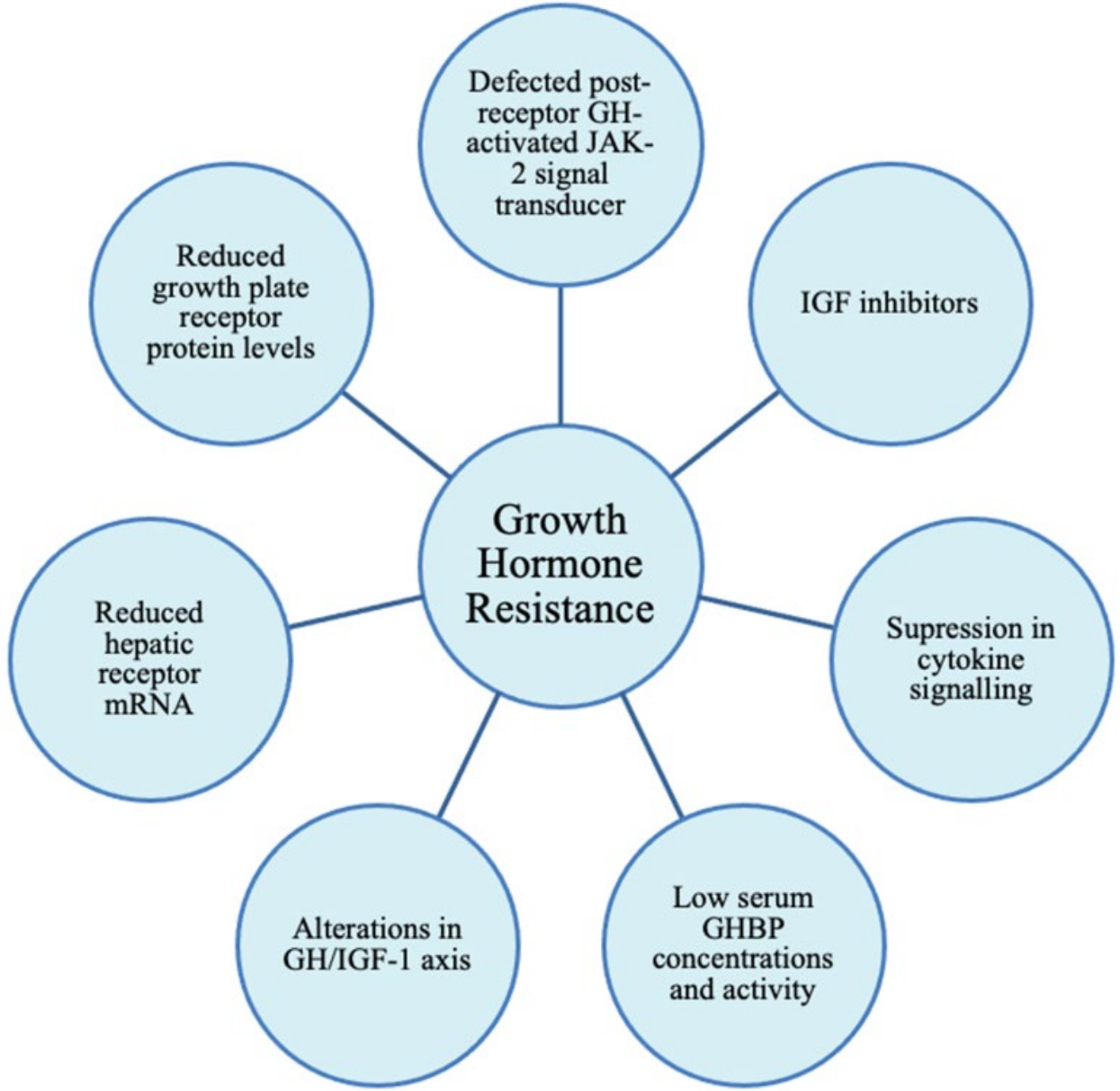
Mechanical effects: Physical inactivity, arthropathy

RAAS activation: Increased angiotensin-2 levels

Testosterone and Vitamin D Deficiencies



Growth Hormone Resistance in CKD



Sarcopenia Management

- Resistance Training
- Vibration Therapy
- Nutritional Supplementation
- Androgen / Androgen Receptor Modulators
- Myostatin Inhibition
- Herbal Supplements

Herbals for Sarcopenia

- *Curcuma longa* – Curcumin
- *Withania somnifera* – Ashwagandha
- *Camellia sinensis* – Green Tea
- *Zingiber officinale* – Ginger
- Grape seeds

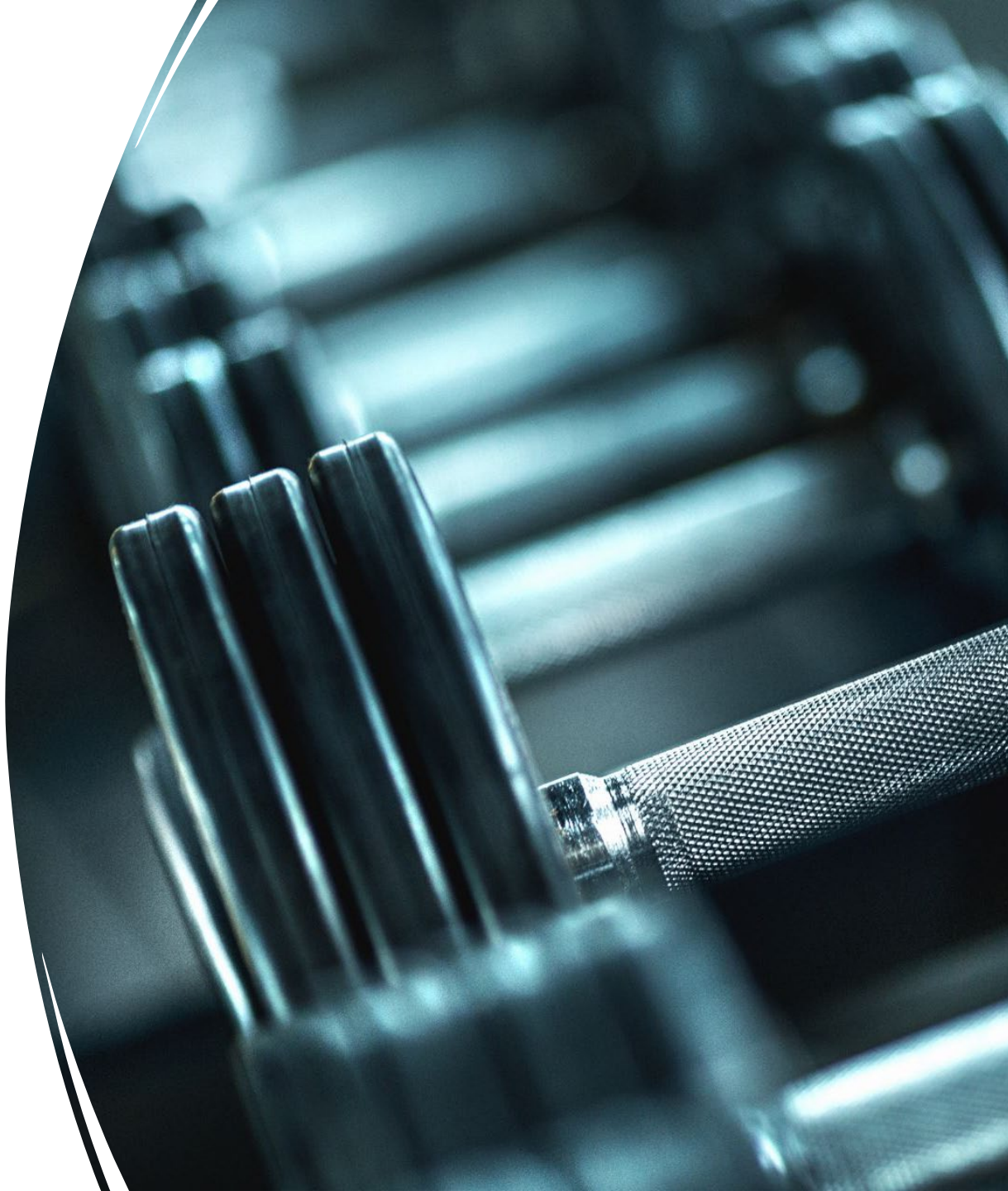
FrOST Trial

- Franconian Osteopenia and Sarcopenia Trial (FrOST)
- Resistance Exercise (RT)
  - HIT – High Intensity Training, low volume
  - DRT – Dynamic Resistance Training
  - Effective in Osteosarcopenia
- 43 sedentary men, age 73-91
  - Dropout, n=2
  - Attendance rate – 95%
  - Unintended side effects / injuries, n=0
  - HIT-RT protocol to be feasible, attractive, and safe

Journal of Bone and Mineral Research, Vol. 35, No. 9, September 2020, pp 1634–1644.

Resistance Training Goals and Outcomes

- Strength vs. Hypertrophy
- Low load - <60% of 1 repetition maximum
- High load - >60% of 1 repetition maximum
- 6 week adaptation period
- Maximal strength – heavy loads
- Muscle hypertrophy – achieved across a spectrum of load ranges



J Strength Cond Res. 2017 Dec;31(12):3508-3523.

Proximity to Failure

- Resistance training to failure vs. non-failure
- Momentary muscular failure does not equate to increased muscle hypertrophy
- Proximity to failure and muscle hypertrophy are not a linear relationship

Sports Medicine (2023) 53:649–665
https://doi.org/10.1007/s40279-022-01784-y

SYSTEMATIC REVIEW

Influence of Resistance Training Proximity-to-Failure on Skeletal Muscle Hypertrophy: A Systematic Review with Meta-analysis

Martin C. Refalo[1] · Eric R. Helms[2] · Eric. T. Trexler[3] · D. Lee Hamilton[4] · Jackson J. Fyfe[4]

Accepted: 16 October 2022 / Published online: 5 November 2022

Abstract

Background and Objective This systematic review with meta-analysis investigated the influence of resistance training proximity-to-failure on muscle hypertrophy.

Methods Literature searches in the PubMed, SCOPUS and SPORTDiscus databases identified a total of 15 studies that measured muscle hypertrophy (in healthy adults of any age and resistance training experience) and compared resistance training performed to: (A) momentary muscular failure versus non-failure; (B) set failure (defined as anything other than momentary muscular failure) versus non-failure; or (C) different velocity loss thresholds.

Results There was a trivial advantage for resistance training performed to set failure versus non-failure for muscle hypertrophy in studies applying any definition of set failure [effect size=0.19 (95% confidence interval 0.00, 0.37), p=0.045], with no moderating effect of volume load (p=0.884) or relative load (p=0.525). Given the variability in set failure definitions applied across studies, sub-group analyses were conducted and found no advantage for either resistance training performed to momentary muscular failure versus non-failure for muscle hypertrophy [effect size=0.12 (95% confidence interval −0.13, 0.37), p=0.343], or for resistance training performed to high (>25%) versus moderate (20–25%) velocity loss thresholds [effect size=0.08 (95% confidence interval −0.16, 0.32), p=0.529].

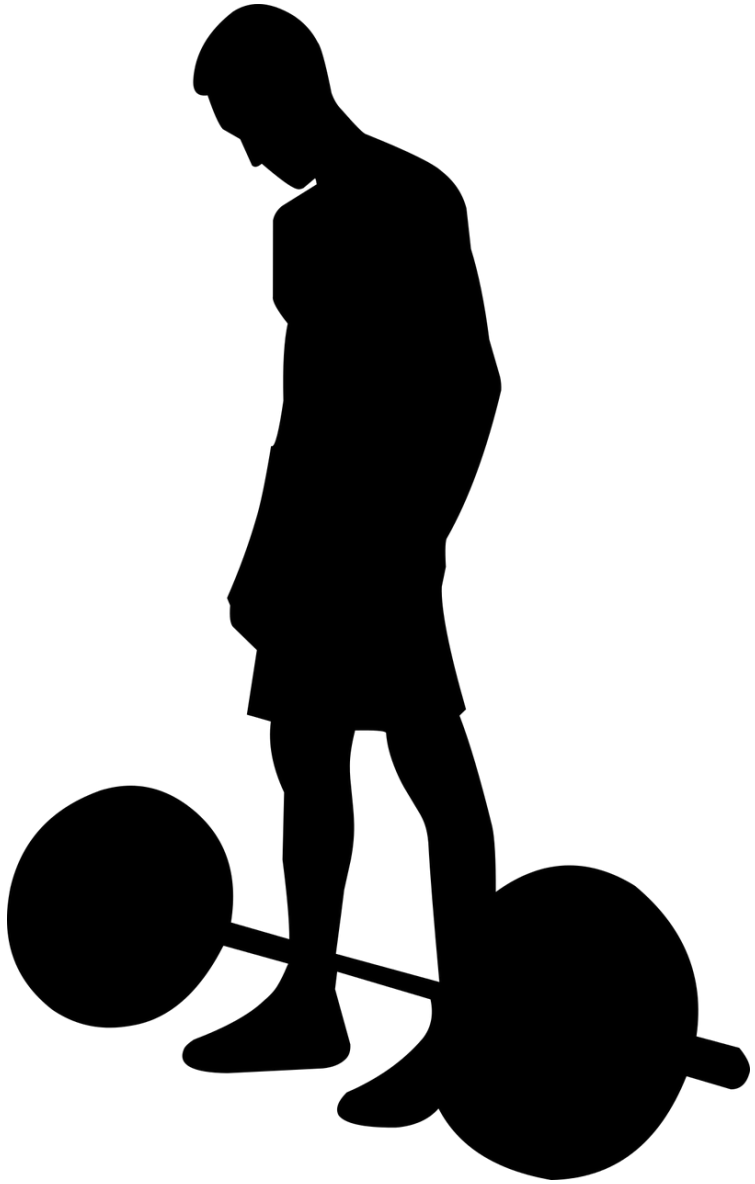
Conclusion Overall, our main findings suggest that (i) there is no evidence to support that resistance training performed to momentary muscular failure is superior to non-failure resistance training for muscle hypertrophy and (ii) higher velocity loss thresholds, and theoretically closer proximities-to-failure do not always elicit greater muscle hypertrophy. As such, these results provide evidence for a potential non-linear relationship between proximity-to-failure and muscle hypertrophy.

Gauging Proximity to Failure

- RPE – Rate of Perceived Effort
  - 1 – Ability to continually perform activity without perceived failure
  - 7 – 3 reps until failure
  - 8 – 2 reps until failure
  - 9 – 1 additional rep could have been completed before failure
  - 10 – Performed until momentary failure

Effective Resistance Exercise

- Easy-to-use
- Avoid complex movements
- Functional resistance training
- Consider activities of daily living
- Emphasize compound exercises
  - Upper body press
  - Upper body pull
  - Overhead press
  - Squat
  - Deadlift



Resistance Training Outside of the Box

- Progression without a high load
  - Necessary in the aging population
  - Blood-flow-restricted-low-load resistance training
    - 20–30% of one repetition maximum (1RM)

(A) H-RT **(B) L-BFR** **(C) L-ST / L-FAIL**

Figure 1. Pictures show a comparison of resistance training of the biceps brachii in each style when using free weights (**A**–**C**). H-RT, high-load (at 70–80% one repetition maximum (1RM)) resistance training. L-BFR, low-load resistance training (at 20–30% 1RM) with blood flow restriction by an elastic designed cuff belt. L-ST, low-load (at 20–30% 1RM) resistance training with relatively slow movement and tonic force generation. L-FAIL, low-load (at 20–30% 1RM) resistance exercise to volitional fatigue.

Designs Schemes

Characteristics	Resistance Training			
	H-RT	L-BFR	L-ST	L-FAIL
Load	70–85% 1RM	10–50% 1RM	BW, 30–50% 1RM	20% 1RM
Frequency (day/week)	2–3	2–3	2–7	3
Sets x Repetitions	1–3 x 8–15	1–4 x 15–30	1–3 x 5–15	1 x 80–100
References	[6,14,17,18]	[9,19–25]	[26–29]	[30]

The numbers in parentheses are references. 1RM, one repetition maximum. BW, body weight. H-RT, high-load resistance training. L-BFR, low-load resistance training with blood flow restriction by an elastic designed cuff belt. L-FAIL, low-load resistance exercise to volitional fatigue. L-ST, low-load resistance training with relatively slow movement and tonic force generation.

	Resistance Training			
	H-RT	L-BFR	L-ST	L-FAIL
Benefits 1	Exercise repetition: Few [6,18,19]	Exercise load: Low [9,19–25]	Exercise load: Low [26,27]	Exercise load: Low [30]
Benefits 2	Strength gain: Large [9,38]	Arterial stiffness: No change [21–23]		
Benefits 3		Versatility: High [9,22,25,61]		
Potential complications 1	Pain in bones and joints: Occurrence [9]	Muscle soreness and damage: Occurrence * [62,63]	Not applicable (Few reports)	Muscle soreness and damage: Occurrence * [64]
Potential complications 2	Arterial stiffness: Increase * [8]	Rhabdomyolysis: Occurrence * [65,66]		Discomfort: Occurrence * [64]
Potential complications 3	Muscle soreness and damage: Occurrence [67]			

The numbers in parentheses are references. * = Reference of young adults. H-RT, high-load (at 70–80% one repetition maximum (1RM)) resistance training. L-BFR, low-load resistance training with blood flow restriction by an elastic designed cuff belt. L-FAIL, low-load resistance exercise to volitional fatigue. L-ST, low-load resistance training with relatively slow movement and tonic force generation.

Cells 2022, 11, 1389.

Low Intensity BFR vs. Low Intensity

- LI-BFR, but not LI improved strength, muscle mass, IGF-1, endothelial function, and selected inflammatory markers in a nonagenarian sarcopenic patient.
- These results are promising and suggest that LI-BFR should be considered as an alternative to prevent muscle loss and improve functional fitness in frail older populations.

Clinical Interventions in Aging

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CASE REPORT

Strength training with blood flow restriction – a novel therapeutic approach for older adults with sarcopenia? A case report

This article was published in the following Dove Press journal:
Clinical Interventions in Aging

Karynne Grutter Lopes[1,2]
Daniel Alexandre Bottino[1,2]
Paulo Farinatti[3–5]
Maria das Graças Coelho de Souza[1,2]
Priscila Alves Maranhão[2]
Clara Maria Soares de Araujo[2]
Eliete Bouskela[1,2]
Roberto Alves Lourenço[6]
Ricardo Brandão de Oliveira[1,5,7]

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Introduction: A 91-year-old sedentary man presenting exhaustion, lower-limb weakness, hypertension, and history of multiple falls was diagnosed with sarcopenia – appendicular skeletal muscle mass index (ASM) of 7.10 kg/m^2.

Purpose: To investigate the effects of strength training performed with low intensity in isolation (LI) or with blood flow restriction (LI-BFR) on strength, muscle mass, IGF-1, endothelial function, microcirculation, inflammatory biomarkers, and oxidative stress.

Methods: In the first 3 months, LI was performed with intensity corresponding to 30% of 1 repetition maximum, followed by 1 month of inactivity, and another 3 months of LI-BFR (similar load than LI concomitant to BFR equivalent to 50% of resting systolic blood pressure).

Results: LI-BFR, but not LI improved muscle mass, ASM, handgrip strength, isokinetic peak torque, IL-6, and IGF-1. Endothelial function, red blood cell velocity, and concentrations of C-reactive protein, and soluble intercellular adhesion molecules-1 improved after both LI and LI-BFR. Endothelin-1 and oxidative stress increased after LI-BFR, and lowered after LI.

Conclusion: LI-BFR, but not LI improved strength, muscle mass, IGF-1, endothelial function, and selected inflammatory markers in a nonagenarian sarcopenic patient. These results are promising and suggest that LI-BFR should be considered as an alternative to prevent muscle loss and improve functional fitness in frail older populations.

Keywords: aging, muscle mass, resistance exercise, vascular occlusion, endothelial function, microcirculation

Bodyweight?

- Only bodyweight resistance exercise with slow movement and plyometric exercise
- Improve physical function in the elderly
- Single sets for each exercise, but no enhanced muscle hypertrophy
- Performing multiple sets required to induce muscle hypertrophy

Geriatr Gerontol Int 2015; 15: 1270-1277.

No Excuses

- Even if the intensity is as low as 30% 1RM, low intensity, slow movement and tonic force generation can increase muscle size and strength in healthy older adults
- Avoiding the burn
  - Training at volitionally very slow durations (>10secs per repetition) is inferior for hypertrophy
- Machines have the ability to reverse sarcopenia
- Progressive resistance training is effective in the oldest of the old

Clin Physiol Funct Imaging. 2014 Nov;34(6):463-70.

Sports Med. 2015 Apr;45(4):577-85.

Experimental Gerontology 163 (2022) 111767

How Old?

J Nutr Health Aging. 2020;24(10):1087-1093

EFFECTIVENESS OF A SHORT-TERM MIXED EXERCISE PROGRAM FOR TREATING SARCOPENIA IN HOSPITALIZED PATIENTS AGED 80 YEARS AND OLDER: A PROSPECTIVE CLINICAL TRIAL

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Abstract: *Objectives:* To assess the effectiveness of short-term exercise for treating sarcopenia in hospitalized older patients aged 80 years and over. *Design:* Prospective clinical trial. *Setting:* A post-acute care unit. *Participants:* Sarcopenic patients aged 80 years or over. *Interventions:* The participants were allocated to the intervention group (to receive a mixed exercise program with 10 sessions over two weeks) or the control group (usual care) based on the sequence of admission. *Outcomes:* The primary outcome was the improvement in activities of daily living (ADL) estimated by the change in Barthel Index (BI) score from the baseline to the end of the 2-week intervention. The secondary outcomes were the changes in gait speed, handgrip strength, the time "UP & GO" test (TUG) score, and the Short Physical Performance Battery (SPPB) score. *Results:* We included

Conclusion: Very old inpatients with sarcopenia can benefit from a mixed exercise program (even as short as two weeks) by improving their ADL and gait speed.

However, the long-term effects of exercise on important clinical outcomes need to be further evaluated.

Key words: Muscle wasting, rehabilitation, post-acute care, activities of daily living.

RT with Type II Sarcopenia

- Sandbag training
  - (0.5 kg at the beginning to 1 kg after 1 month)
- Training group
  - Significant improvements
    - Glycosylated hemoglobin
    - 5 times sit-to-stand test
    - Skeletal muscle mass
    - Calf circumference
    - Sarcopenia questionnaire

International Journal of *Environmental Research and Public Health*

MDPI

Article

Effects of 12-Week Progressive Sandbag Exercise Training on Glycemic Control and Muscle Strength in Patients with Type 2 Diabetes Mellitus Combined with Possible Sarcopenia

Yu-Hsuan Chien [1,2], Chia-Jen Tsai [1], Dean-Chuan Wang [2,3], Pin-Hung Chuang [4] and Hwai-Ting Lin [2,3,*]

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Abstract: Patients with type 2 diabetes mellitus (T2DM) are at a three-fold increased risk of developing sarcopenia compared to those without diabetes. The objective of this study was to investigate whether an intervention involving progressive sandbag exercises is beneficial to patients with T2DM and possible sarcopenia in terms of enhancing muscle strength and controlling blood sugar levels. Forty patients with T2DM and possible sarcopenia (age > 50 years) were recruited and randomly divided into resistance training and control groups. Resistance exercises for the upper and lower extremities were performed using sandbags (0.5 kg at the beginning to 1 kg after 1 month). Patients in the control group were asked to maintain their usual daily lifestyle. After 12 weeks, the training group were significant better than the control group in terms of glycosylated hemoglobin, the five times sit-to-stand test, skeletal muscle mass and calf circumference, and the physiological domain of the World Health Organization Quality of Life Questionnaire. In conclusion, these simple home exercises are beneficial to patients with T2DM combined with possible sarcopenia. This approach can assist patients in controlling their levels of glycosylated hemoglobin as well as improve physical fitness and quality of life.

check for updates

Citation: Chien, Y.-H.; Tsai, C.-J.; Wang, D.-C.; Chuang, P.-H.; Lin, H.-T. Effects of 12-Week Progressive Sandbag Exercise Training on Glycemic Control and Muscle Strength in Patients with Type 2 Diabetes Mellitus Combined with Possible Sarcopenia. *Int. J. Environ. Res. Public Health* **2022**, *19*, 15009. https://doi.org/10.3390/ijerph192215009

Programming Workouts

- Consider volume first
  - Volume capabilities decline with age
  - Slower recovery ability
- 3-4 days per week
- Split body movements
- Limit to 2-3 working sets
- Stay within a 30-45 window
- Intensity is relative, use RPE scale (Optimal 60-85%)
- Deadlifting is the most taxing
- Group / social setting if possible (individual variability)

Exercise Dosage Based on Objectives

Effects and examples of recommended training dosages and possible organizational approaches to different forms of strength training for elderly people

Objectives	Possible effects of training	Dosage	Possible organizational approaches
Increase in muscle strength	Increase in muscle mass	8–12 repetitions per muscle group in 70–85 % of the one-repetition-maximum, 3 sets; 2–3 training units per week; at least 8-12 weeks	Fitness studio; gymnasium, home program, initially under instruction, later independently
	Training of intramuscular coordination	Up to 8 repetitions per muscle group with intensities of more than 80% of the one-repetition-maximum; 3–5 sets; 3 training units per week; several weeks	Fitness studio; gymnasium, home program, under instruction
	Training of intermuscular coordination	Several repetitions; up to daily training units; high speed of movement, among others	Training on uneven surfaces with or without additional weights; under instruction, later independently
Reduction of sarcopenia	Increase in muscle mass	8–12 repetitions per muscle group in 60–80% of the one-repetition-maximum; 3 sets, 3 training units per week, at least 8–12 weeks	Fitness studio; gymnasium, home program, initially under instruction, later independently
Adaptation of tendons and bones	Increase in net synthesis of collagen; reduction in bone density loss	Medium to high intensities (>60–80% of the one-repetition-maximum, >body weight); several training units per week; weeks to months	Fitness studio; gymnasium, under instruction
Prevention of falls and injuries	Optimizing postural control; training of intermuscular coordination	Several repetitions; up to daily training units; high speed of movement	Training on uneven surfaces with or without additional weights; under instruction, later independently
	Training of intramuscular coordination	Up to 8 repetitions per muscle group in intensities of more than 80% of the one-repetition-maximum; 3–5 sets; 3 training units per week; several weeks	Fitness studio; gymnasium, home program, under instruction

Dtsch Arztebl Int 2011; 108(21): 359–64.

Gut-Muscle Axis

- Gut microbiota acts as a nutritional mediator of muscle cells
  - Cross-road between nutrition and muscle function
- Gut microbiota
  - As much as 10^{14} bacteria, viruses, fungi, protozoa, and Archaea
  - Gene pool 150 times larger than that of the host
  - Weight esteemed between 175 g and 1.5 kg (3.3 lb)
  - 1100 and 2000 bacterial taxa
  - Maturity by age 3
  - Stable over the lifespan
  - 10 phyla
  - Two phyla—*Bacteroidetes* and *Firmicutes*—account for as much as 99% of species
- Influences physical performance and muscle function through CNS

Gut Microbiota in Aging

After age 65, resilience is generally reduced

Composition more vulnerable to lifestyle changes, antibiotics and disease

Biodiversity declines

Dysbiosis is correlated with physical frailty

Reduced *Faecalibacterium prausnitzii* in frail patients

- Reduced SCFA production and metabolic modulation

Gut microbiome is involved in aging phenotype and in the onset of physical frailty

Exercise Influence on the Gut Microbiota

- Over-representation of some health-promoting bacterial species, including *Akkermansia, Faecalibacterium*, and *Roseburia*
- Enhanced biodiversity
- Correlation with cardiorespiratory fitness
- “Transducer” of nutrient signals for host
- Produces mediators that influence metabolic balance, insulin sensitivity, and inflammation
- Healthy gut microbiota – anabolism
- Dysbiotic microbiota – anabolic resistance, or even catabolism

Muscle Effects of Microbial Metabolites

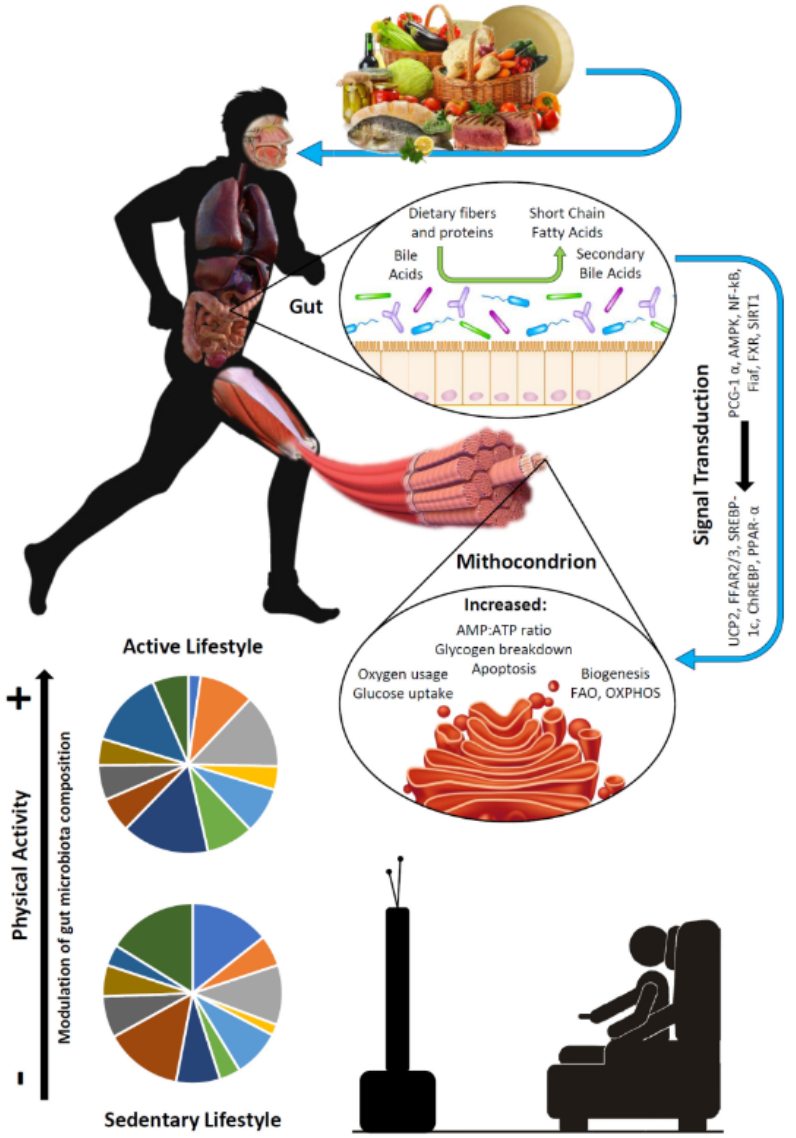
Table 3. Overview of the main microbial metabolites acting as nutrients or metabolic/physiological modulators for the host, which are also possibly involved in skeletal muscle function.

Substance	Bacterial Taxa Involved	Possible Effects on Muscle
Folate	*Bifidobacteria* *lactobacilli*	Biosynthesis of amino acids DNA synthesis, methylation, and repair
Riboflavin (vitamin B_2)	*Bacillus subtilis* *Escherichia coli* *Bifidobacteria*	Improvement of redox reactions and energy production Improved resistance to fatigue
Vitamin B_{12}	*Propionibacteria* *Lactobacillus reuteri*	Preservation of strength through the prevention of homocysteine-induced oxidative stress and endothelial damage
Glycine betaine	*Escherichia coli* *Klebsiella*	Stimulation of anabolism and cell proliferation by IGF-1 synthesis
Tryptophan	Several bacterial species	Stimulation of anabolism and cell proliferation by IGF-1 synthesis
Short-chain fatty acids	*Faecalibacterium* *Butyricimonas* *Succinivibrio* *Pseudosuccinivibrio*	Promotion of insulin sensitivity, modulation of inflammation, promotion of mitochondrial biogenesis, and energy production
Urolithins	Several bacterial species involved (not fully identified)	Preservation of skeletal muscle cell mitochondrial biogenesis and activity, promotion of muscle anabolism

IGF-1: insulin-like growth factor-1.

Cross-Road Between Nutrition and Muscle

- Diet influences microbiota
- Microbiota metabolizes nutrients into mediators
- Mediators influence myocytes
  - Especially mitochondria
- Modulate inflammation
- Promotes insulin sensitivity



Nutrients 2017, 9, 1303

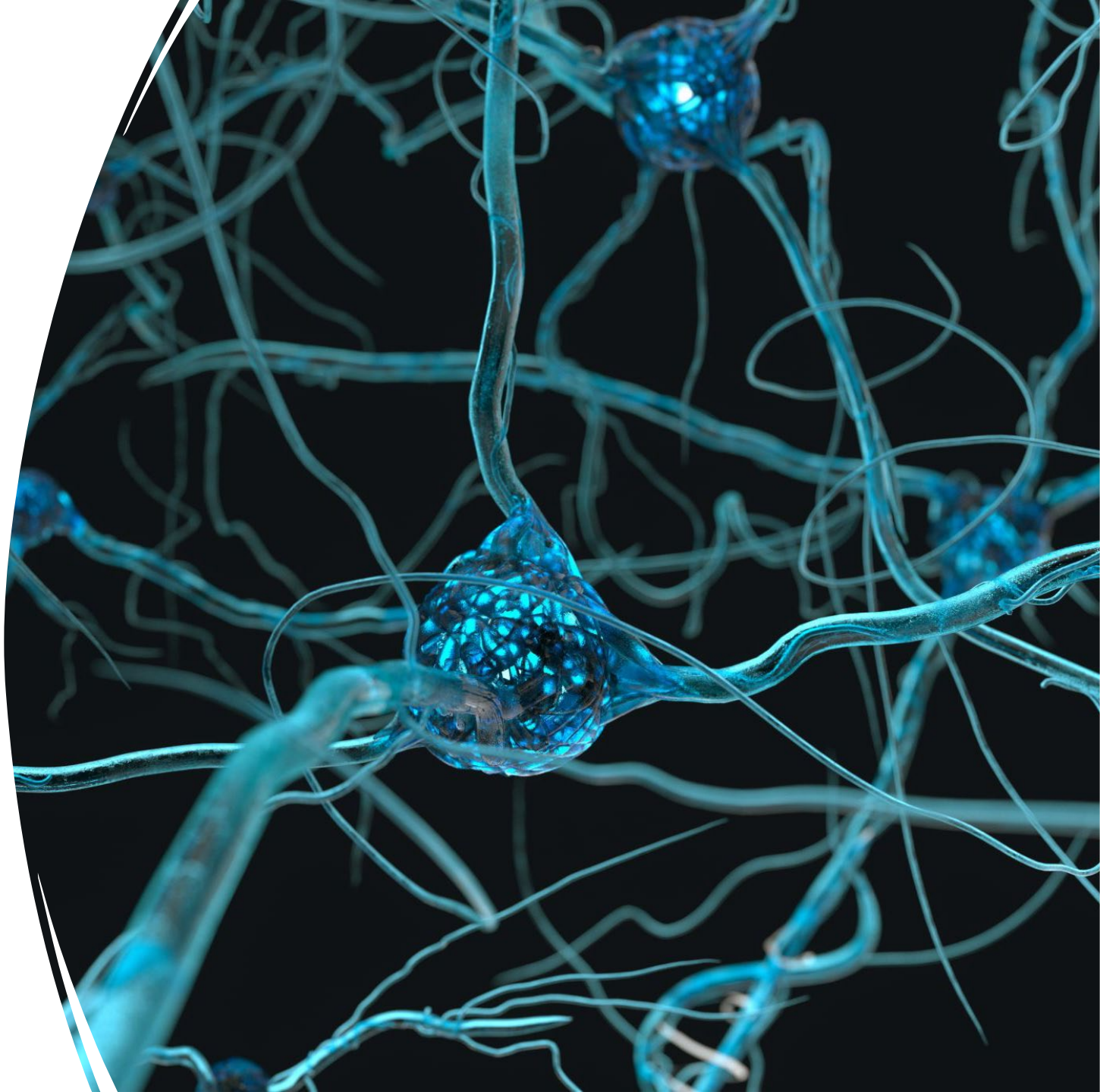
Physical Activity and Neurodegeneration: Role of Muscle-Brain Axis

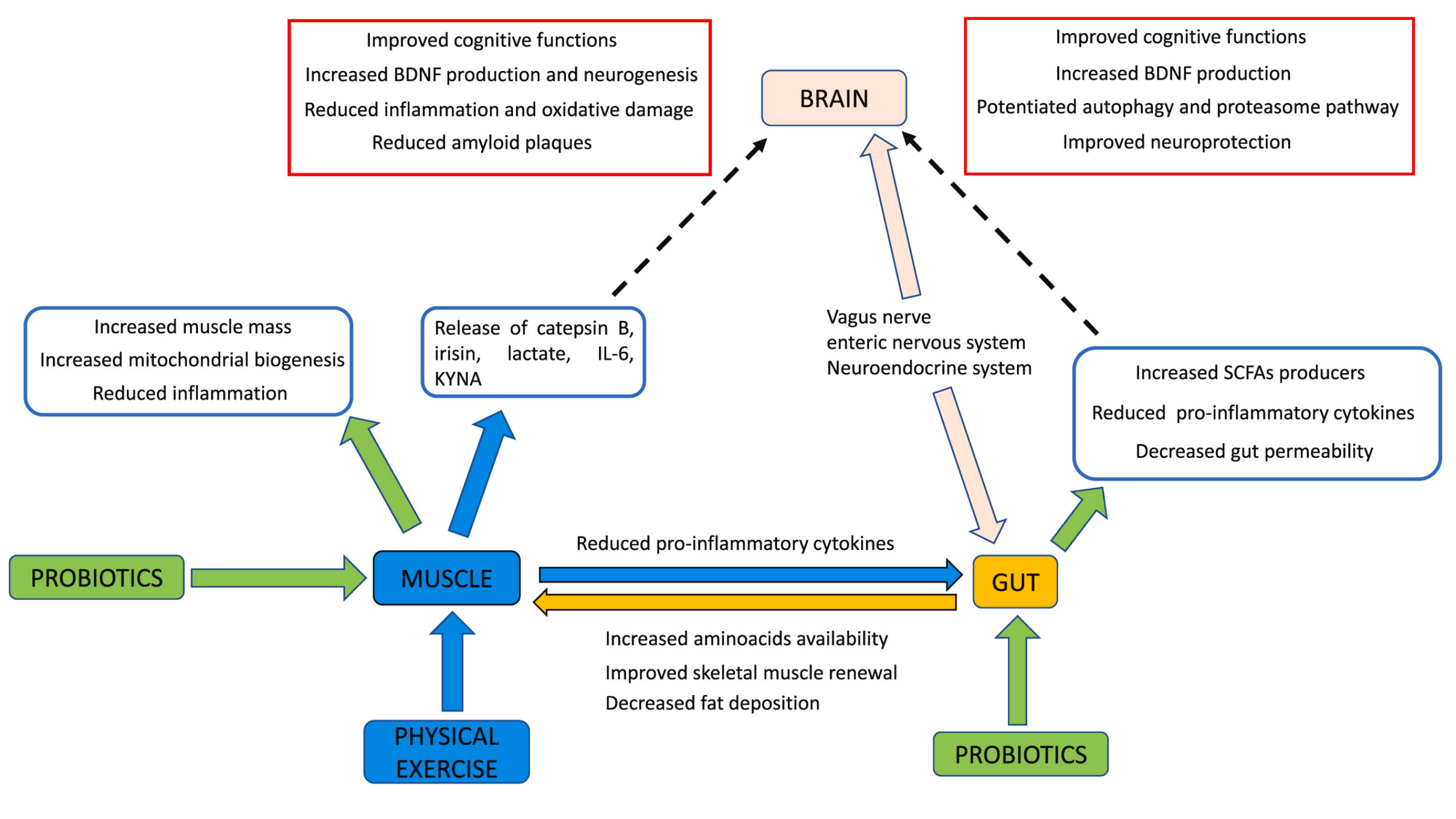
- Increase in synaptic plasticity
- Reduces accumulation of amyloid plaques
- Upregulation of SIRT-1
- Increased BDNF (brain derived neurotrophic factor)
- Reduction in hyperphosphorylated Tau
- Decreased activation of microglia
- Decreased risk of all-cause dementia, even at 20 year follow up
- Decreased neuroinflammatory markers
- Improved mitochondrial function
- Aids in motor symptoms (Parkinson's)

Neuropharmacology 240 (2023) 109718

Probiotic Supplementation

- Bifidobacterium, Lactobacillus and *Streptococcus thermophilus*
- Reduced anxiety
- Reduced accumulation of amyloid
- Restoration of neuronal proteolytic pathways
  - Ubiquitin proteasome pathway
  - Autophagy
- Reduced oxidative stress
- 12 week intervention period is common




Improved cognitive functions
Increased BDNF production and neurogenesis
Reduced inflammation and oxidative damage
Reduced amyloid plaques
BRAIN
Improved cognitive functions
Increased BDNF production
Potentiated autophagy and proteasome pathway
Improved neuroprotection
Increased muscle mass
Increased mitochondrial biogenesis
Reduced inflammation
Release of catepsin B, irisin, lactate, IL-6, KYNA
Vagus nerve
enteric nervous system
Neuroendocrine system
Increased SCFAs producers
Reduced pro-inflammatory cytokines
Decreased gut permeability
PROBIOTICS
MUSCLE
Reduced pro-inflammatory cytokines
GUT
Increased aminoacids availability
Improved skeletal muscle renewal
Decreased fat deposition
PHYSICAL EXERCISE
PROBIOTICS

Overlap

- Considering the reciprocal influence of gut microbiota and muscle physiology, probiotic supplementation may represent a novel strategy to optimize muscle functionality and metabolism in either anabolic (exercise) or catabolic (sarcopenia and cachexia) conditions.
- Considering the influence of both gut microbiota and the muscle on brain functions and the crosstalk occurring between these systems, it is reasonable to hypothesize a synergistic effect mediated by a combined intervention with probiotics and exercise training.

Sarcopenia and Nutrition

- Age-related loss of appetite
- “Anorexia of aging”
- Increased energy requirements due to acute and chronic inflammation
- “Disease-related malnutrition”
- Typical prescriptions
  - Adequate intakes of proteins
  - Vitamin D
  - Antioxidant nutrients
  - Long-chain polyunsaturated fatty acids

Sarcopenia and Nutrition – What We Know

Macronutrients

- Protein
  - Lower intake related to loss of lean mass by DXA and reduced grip strength
  - Improved appendicular muscle mass
  - Leucine increases muscle protein synthesis in older individuals

Micronutrients

- Vitamin D
  - Improved muscle mass, not grip strength
  - Results seen with D3, not D2
- Vitamin C
  - Reduce oxidative stress of sarcopenia
  - Improved physical performance (gait, chair-rise, balance test)

Sarcopenia and Nutrition – What We Know

- Micronutrients
  - B6 (abundant in meat and poultry)
    - Deficiency symptoms – neurological, affect on motor neurons, weakness, loss of distal sensation
    - Improved Short Physical Performance Battery
    - Upregulates several protective myogenic genes
  - B12
    - Deficiency symptoms – neuromuscular, muscle weakness, paresthesia, numbness
    - Greater intake in non-sarcopenic groups
  - Other vitamins
    - Vitamin A, carotenoids – antioxidative
    - Vitamin E - antioxidative

Sarcopenia and Nutrition – What We Know

- Micronutrients (minerals)
  - Calcium – structural and regulatory muscle functions, myogenesis
  - Selenium – selenoproteins, deficiency linked to muscle pain and weakness
  - Magnesium – 600 enzymatic reactions, energy and protein synthesis
  - Phosphorus – preservation of muscle mass
  - Iron – physical function decline with deficiency
  - Zinc – protein synthesis
- Fatty Acids
  - Omega-3s – positive effect on muscle mass, increased protein synthesis

Nutrients 2020, 12, 1755

JAMDA 24 (2023) 1163-1172

Whole Foods

Whole Foods	Muscle Health Outcome
Meats	
	mid-arm muscle area
	ASM index
	SMM
Fruit and vegetables	
	lean muscle mass
	walking speed
	lean muscle mass
	walking speed
	grip strength
	sarcopenia
	physical performance (SFT)

Supplemented LOV [1]	
	muscle strength (upper & lower body)
	muscle power
	muscle size
Dairy products	
	total ASM
	lean body mass in arms
	grip strength
	lean tissue in legs
	total muscle mass
Eggs	
	lean body mass (trunk and total)

Nutrients 2020, 12, 2257;

Milk

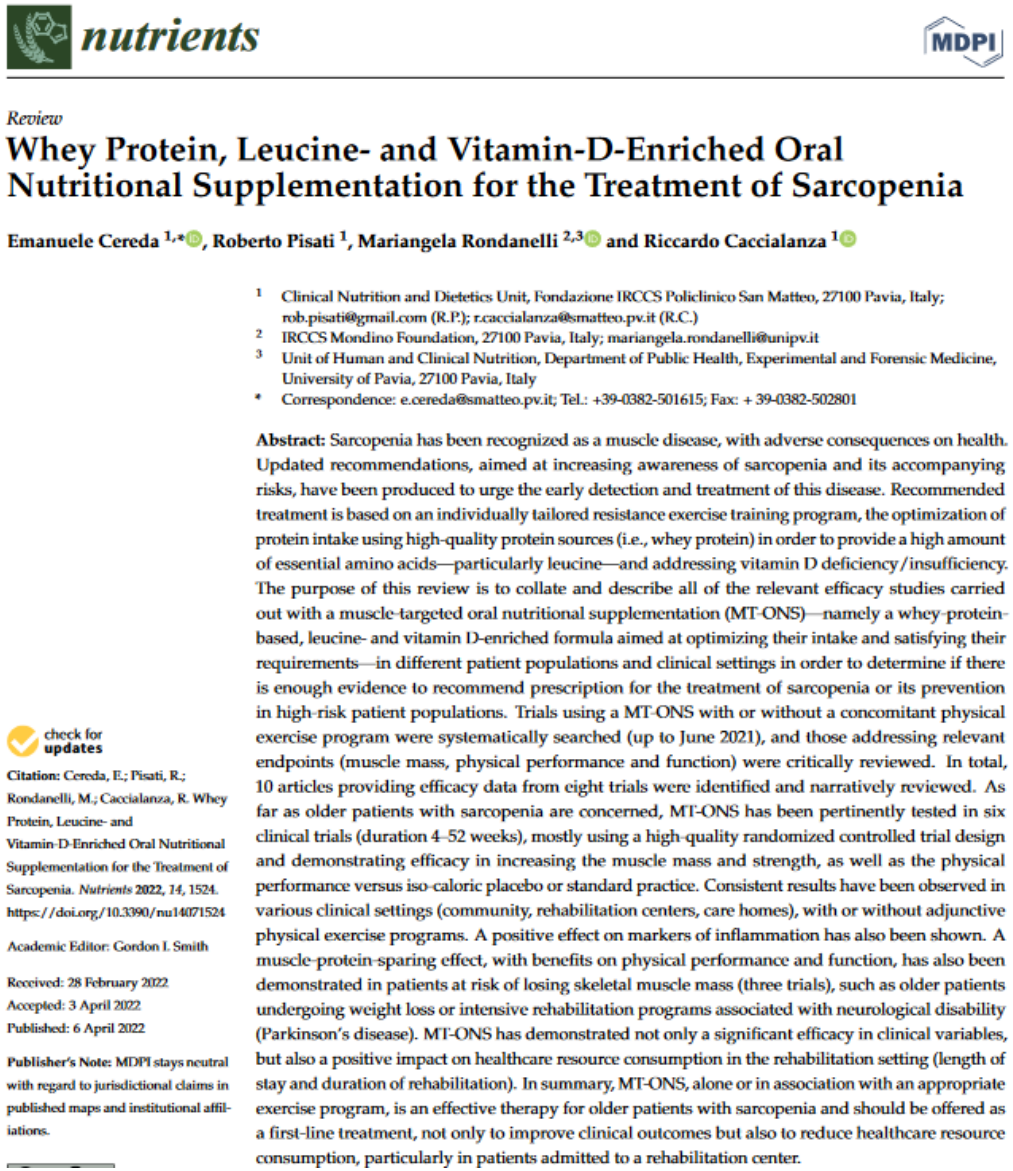
- Contains multiple nutrients and bioactive compounds
- Proteins for anabolism
- Myoprotective
- Bioactive peptides
- Antihypertensive, antithrombotic, antimicrobial and immunomodulatory effects
- Lactoferrin and immunoglobulins

- High-quality proteins (20% of whey and 80% of caseins)
- Minerals (e.g. calcium, phosphorus, magnesium, iodine)
- Vitamins (e.g. fat-soluble A and E and water-soluble B vitamins)
- Carbohydrates (lactose and oligosaccharides)
- Fats—a mixture of 70% saturated (SFA), and 30% of mono- (MUFA) and polyunsaturated fatty acids (PUFA)
- Multiple micronutrients

Clinical Interventions in Aging 2020:15 695–714

Whey Protein, Leucine and Vitamin D

In summary, muscle-targeted oral nutritional supplementation, alone or in association with an appropriate exercise program, is an effective therapy for older patients with sarcopenia and should be offered as a first-line treatment.



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Review

Whey Protein, Leucine- and Vitamin-D-Enriched Oral Nutritional Supplementation for the Treatment of Sarcopenia

Emanuele Cereda [1,*], Roberto Pisati [1], Mariangela Rondanelli [2,3] and Riccardo Caccialanza [1]

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Abstract: Sarcopenia has been recognized as a muscle disease, with adverse consequences on health. Updated recommendations, aimed at increasing awareness of sarcopenia and its accompanying risks, have been produced to urge the early detection and treatment of this disease. Recommended treatment is based on an individually tailored resistance exercise training program, the optimization of protein intake using high-quality protein sources (i.e., whey protein) in order to provide a high amount of essential amino acids—particularly leucine—and addressing vitamin D deficiency/insufficiency. The purpose of this review is to collate and describe all of the relevant efficacy studies carried out with a muscle-targeted oral nutritional supplementation (MT-ONS)—namely a whey-protein-based, leucine- and vitamin D-enriched formula aimed at optimizing their intake and satisfying their requirements—in different patient populations and clinical settings in order to determine if there is enough evidence to recommend prescription for the treatment of sarcopenia or its prevention in high-risk patient populations. Trials using a MT-ONS with or without a concomitant physical exercise program were systematically searched (up to June 2021), and those addressing relevant endpoints (muscle mass, physical performance and function) were critically reviewed. In total, 10 articles providing efficacy data from eight trials were identified and narratively reviewed. As far as older patients with sarcopenia are concerned, MT-ONS has been pertinently tested in six clinical trials (duration 4–52 weeks), mostly using a high-quality randomized controlled trial design and demonstrating efficacy in increasing the muscle mass and strength, as well as the physical performance versus iso-caloric placebo or standard practice. Consistent results have been observed in various clinical settings (community, rehabilitation centers, care homes), with or without adjunctive physical exercise programs. A positive effect on markers of inflammation has also been shown. A muscle-protein-sparing effect, with benefits on physical performance and function, has also been demonstrated in patients at risk of losing skeletal muscle mass (three trials), such as older patients undergoing weight loss or intensive rehabilitation programs associated with neurological disability (Parkinson's disease). MT-ONS has demonstrated not only a significant efficacy in clinical variables, but also a positive impact on healthcare resource consumption in the rehabilitation setting (length of stay and duration of rehabilitation). In summary, MT-ONS, alone or in association with an appropriate exercise program, is an effective therapy for older patients with sarcopenia and should be offered as a first-line treatment, not only to improve clinical outcomes but also to reduce healthcare resource consumption, particularly in patients admitted to a rehabilitation center.

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Citation: Cereda, E.; Pisati, R.; Rondanelli, M.; Caccialanza, R. Whey Protein, Leucine- and Vitamin-D-Enriched Oral Nutritional Supplementation for the Treatment of Sarcopenia. *Nutrients* **2022**, *14*, 1524. https://doi.org/10.3390/nu14071524

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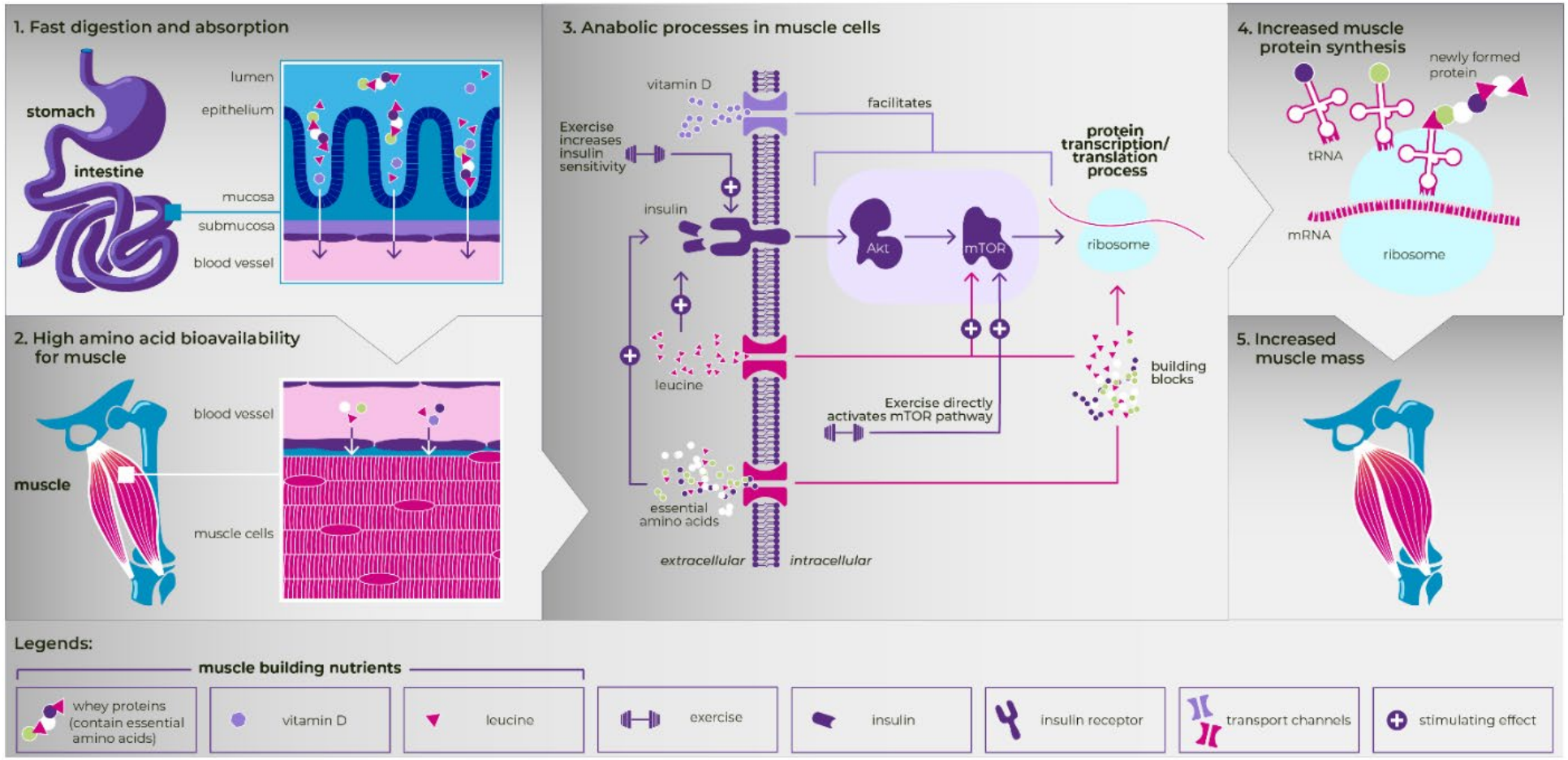
Whey Protein, Leucine- and Vitamin-D



Protein

The Most Dynamic, but Underused Macronutrient

Protein & Sarcopenia

- Protein impacts the microbiota
- Associations between red meat proteins and altered gut microbiota
  - Caution: Observational studies, not interventional
- Combination of plant and animal-based proteins are deemed favorable for gut microbiota eubiosis and muscle-protein synthesis
  - Highlights the role of fiber to enhance microbial activity and SCFA
  - Insufficient consideration for polyphenol / secondary metabolite concentrations in appropriately raised animal foods
  - Consideration for digestive capacity and if >10% protein reaches the large intestine
- High-protein diets with elevated essential amino-acid concentrations, alongside increased dietary fiber intake, may promote gut microbiota eubiosis
  - Metabolic effects derived from short-chain fatty-acid and branched-chain fatty-acid production

Long Term Experimental Trials

- Data on high proteins diets and the gut microbiota
- However, to date, there is no causal link in humans, taking into account the absence of long-term experimental trials on high protein diets and the gut microbiome and their multifaceted relationships.
- Further controversy
  - Multiple human and animal studies have linked increased branched-chain amino acids (BCAAs) with insulin resistance and type 2 diabetes in obese groups. However, increased SCFA consumption may alleviate the hyperglycemic responses that are occurring in obese and type 2 diabetics, characterized by elevated amino-acid concentrations

Nutrients 2020, 12, 2285

Animal vs. Plant Protein – Anabolic Properties

- Plant-based proteins
  - Less anabolic effect
  - Lower digestibility
  - Lower essential amino acid content (especially leucine)
  - Deficient in other essential amino acids (sulfur amino acids or lysine)
  - Directed toward oxidation rather than muscle protein synthesis
- Improving amino acid composition
  - Fortification with amino acids (leucine)
  - Selective breeding
  - Blending plant protein sources
  - Blending with animal protein sources
  - Greater dose of plant protein intake
  - Leucine co-ingestion

Nutrients 2019, 11, 1825

Proc Nutr Soc. 2018 Feb;77(1):20-31.

J Nutr. 2015 Sep;145(9):1981-91.

Additional Considerations for Aging

- High intake of plant-based protein-rich products
  - Could pose risks of malnutrition
  - Fiber-related intestinal intolerances
- Lacks the known bioactive peptides found in animal proteins
  - Lactoferrin

Curr Opin Clin Nutr Metab Care. 2024 Jul 1;27(4):372-377.

Lactoferrin Plus

- Regarding muscle function, all the tests significantly improved in the supplemented group compared to the placebo one.
- CRP, zonulin, and TNF-alpha significantly decreased in intervention, compared to placebo.
- This dietary supplement improves muscle mass and function, and the gut muscle has emerged as a new intervention target for sarcopenia.

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Article

A Patented Dietary Supplement (Hydroxy-Methyl-Butyrate, Carnosine, Magnesium, Butyrate, Lactoferrin) Is a Promising Therapeutic Target for Age-Related Sarcopenia through the Regulation of Gut Permeability: A Randomized Controlled Trial

Mariangela Rondanelli [1], Clara Gasparri [2,*], Alessandro Cavioni [2], Claudia Sivieri [2], Gaetan Claude Barrile [2], Francesca Mansueto [2] and Simone Perna [3]

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Citation: Rondanelli, M.; Gasparri, C.; Cavioni, A.; Sivieri, C.; Barrile, G.C.; Mansueto, F.; Perna, S. A Patented Dietary Supplement (Hydroxy-Methyl-Butyrate, Carnosine, Magnesium, Butyrate, Lactoferrin) Is a Promising Therapeutic Target for Age-Related Sarcopenia through the Regulation of Gut Permeability: A Randomized Controlled Trial. *Nutrients* **2024**, *16*, 1369. https://doi.org/10.3390/nu16091369

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Revised: 26 April 2024

Abstract: Adequate diet, physical activity, and dietary supplementation with muscle-targeted food for special medical purposes (FSMP) or dietary supplement (DS) are currently considered fundamental pillars in sarcopenia treatment. The aim of this study is to evaluate the effectiveness of a DS (containing hydroxy-methyl-butyrate, carnosine, and magnesium, for its action on muscle function and protein synthesis and butyrate and lactoferrin for their contribution to the regulation of gut permeability and antioxidant/anti-inflammation activity) on muscle mass (assessed by dual X-ray absorptiometry (DXA)), muscle function (by handgrip test, chair test, short physical performance battery (SPPB) test, and walking speed test), inflammation (tumor necrosis factor-alpha (TNF-a), C-reactive protein (CRP), and visceral adipose tissue (VAT)) and gut axis (by zonulin). A total of 59 participants (age 79.7 ± 4.8 years, body mass index 20.99 ± 2.12 kg/m^2) were enrolled and randomly assigned to intervention (n = 30) or placebo (n = 28). The skeletal muscle index (SMI) significantly improved in the supplemented group compared to the placebo one, +1.02 (CI 95%: −0.77; 1.26), p = 0.001; a significant reduction in VAT was observed in the intervention group, −70.91 g (−13.13; −4.70), p = 0.036. Regarding muscle function, all the tests significantly improved (p = 0.001) in the supplemented group compared to the placebo one. CRP, zonulin, and TNF-alpha significantly decreased (p = 0.001) in intervention, compared to placebo, −0.74 mg/dL (CI 95%: −1.30; −0.18), −0.30 ng/mL (CI 95%: −0.37; −0.23), −6.45 pg/mL (CI 95%: −8.71; −4.18), respectively. This DS improves muscle mass and function, and the gut muscle has emerged as a new intervention target for sarcopenia.

Sarcopenia and Whey Protein

- We consider HIT-RT supported by whey protein supplementation as a feasible, attractive, safe and highly effective option to fight osteosarcopenia in older men.
- 1.5-1.6 gm/kg body mass/day

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Article

Effects of High Intensity Dynamic Resistance Exercise and Whey Protein Supplements on Osteosarcopenia in Older Men with Low Bone and Muscle Mass. Final Results of the Randomized Controlled FrOST Study

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Abstract: The present study aimed to evaluate the effect of high intensity dynamic resistance exercise (HIT-DRT) and whey protein supplementation (WPS) on bone mineral density (BMD) and sarcopenia parameters in osteosarcopenic men. Men ≥ 72 years with osteosarcopenia ($n = 43$) were randomly assigned to a HIT-RT (HIT-RT: $n = 21$) or a non-training control group ($n = 22$). Supervised HIT-RT twice/week was applied for 18 months, while the control group maintained their habitual lifestyle. Supplying WPS, total protein intake amounted to 1.5–1.6 (HIT-RT) and 1.2 g/kg/body mass/d (control). Both groups were supplied with calcium and vitamin D. Primary study outcomes were BMD and the sarcopenia Z-score. After adjusting for multiplicity, we observed significant positive effects for sarcopenia Z-score (standardized mean difference (SMD): 1.40), BMD at lumbar spine (SMD: 0.72) and total hip (SMD: 0.72). In detail, effect sizes for skeletal muscle mass changes were very pronounced (1.97, $p < 0.001$), while effects for functional sarcopenia parameters were moderate (0.87, $p = 0.008$; handgrip strength) or low (0.39, $p = 0.209$; gait velocity). Apart from one man who reported short periods of temporary worsening of existing joint pain, no HIT-RT/WPS-related adverse effects or injuries were reported. We consider HIT-RT supported by whey protein supplementation as a feasible, attractive, safe and highly effective option to fight osteosarcopenia in older men.

Sarcopenia and Creatine

In conclusion, older females supplementing with Creatine experience significant gains in muscle strength, especially when RT lasts for at least 24 weeks in duration.

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Review

Efficacy of Creatine Supplementation Combined with Resistance Training on Muscle Strength and Muscle Mass in Older Females: A Systematic Review and Meta-Analysis

Ellem Eduarda Pinheiro dos Santos [1], Rodrigo Cappato de Araújo [2], Darren G. Candow [3], Scott C. Forbes [4,*], Jaddy Antunes Guijo [1], Carla Caroliny de Almeida Santana [5], Wagner Luiz do Prado [6] and João Paulo Botero [7,*]

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Citation: dos Santos, E.E.P.; de Araújo, R.C.; Candow, D.G.; Forbes, S.C.; Guijo, J.A.; de Almeida Santana, C.C.; Prado, W.L.d.; Botero, J.P. Efficacy of Creatine Supplementation Combined with Resistance Training on Muscle Strength and Muscle Mass in Older Females: A Systematic Review and Meta-Analysis. *Nutrients* **2021**, *13*, 3757. https://doi.org/10.3390/nu13113757

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Abstract: Sarcopenia refers to the age-related loss of muscle strength and muscle mass, which is associated with a reduced quality of life, particularly in older females. Resistance training (RT) is well established to be an effective intervention to counter indices of sarcopenia. Accumulating research indicates that the addition of creatine supplementation (Cr) to RT augments gains in muscle strength and muscle mass, compared to RT alone. However, some evidence indicates that sex differences may alter the effectiveness of Cr. Therefore, we systematically reviewed randomized controlled trials (RCTs) investigating the efficacy of Cr + RT on measures of upper- and lower-body strength and muscle mass in older females. A systematic literature search was performed in nine electronic databases. Ten RCTs (N = 211 participants) were included the review. Overall, Cr significantly increased measures of upper-body strength (7 studies, $n = 142$, $p = 0.04$), with no effect on lower-body strength or measures of muscle mass. Sub-analyses revealed that both upper-body (4 studies, $n = 97$, $p = 0.05$) and lower-body strength (4 studies, $n = 100$, $p = 0.03$) were increased by Cr, compared to placebo in studies $\geq$ 24 weeks in duration. In conclusion, older females supplementing with Cr experience significant gains in muscle strength, especially when RT lasts for at least 24 weeks in duration. However, given the level of evidence, future high-quality studies are needed to confirm these findings.

An Effective Anti-Aging Protocol

A Simplistic Approach to Aging – Stick to the Fundamentals

- Genomic instability
- Telomere attrition
- Epigenetic alterations
- Loss of proteostasis
- Disabled macroautophagy
- Deregulated nutrient-sensing
- Mitochondrial dysfunction
- Cellular senescence
- Stem cell exhaustion
- Altered intercellular communication
- Chronic inflammation
- Dysbiosis

Most Patients Will Benefit From Just the Basics

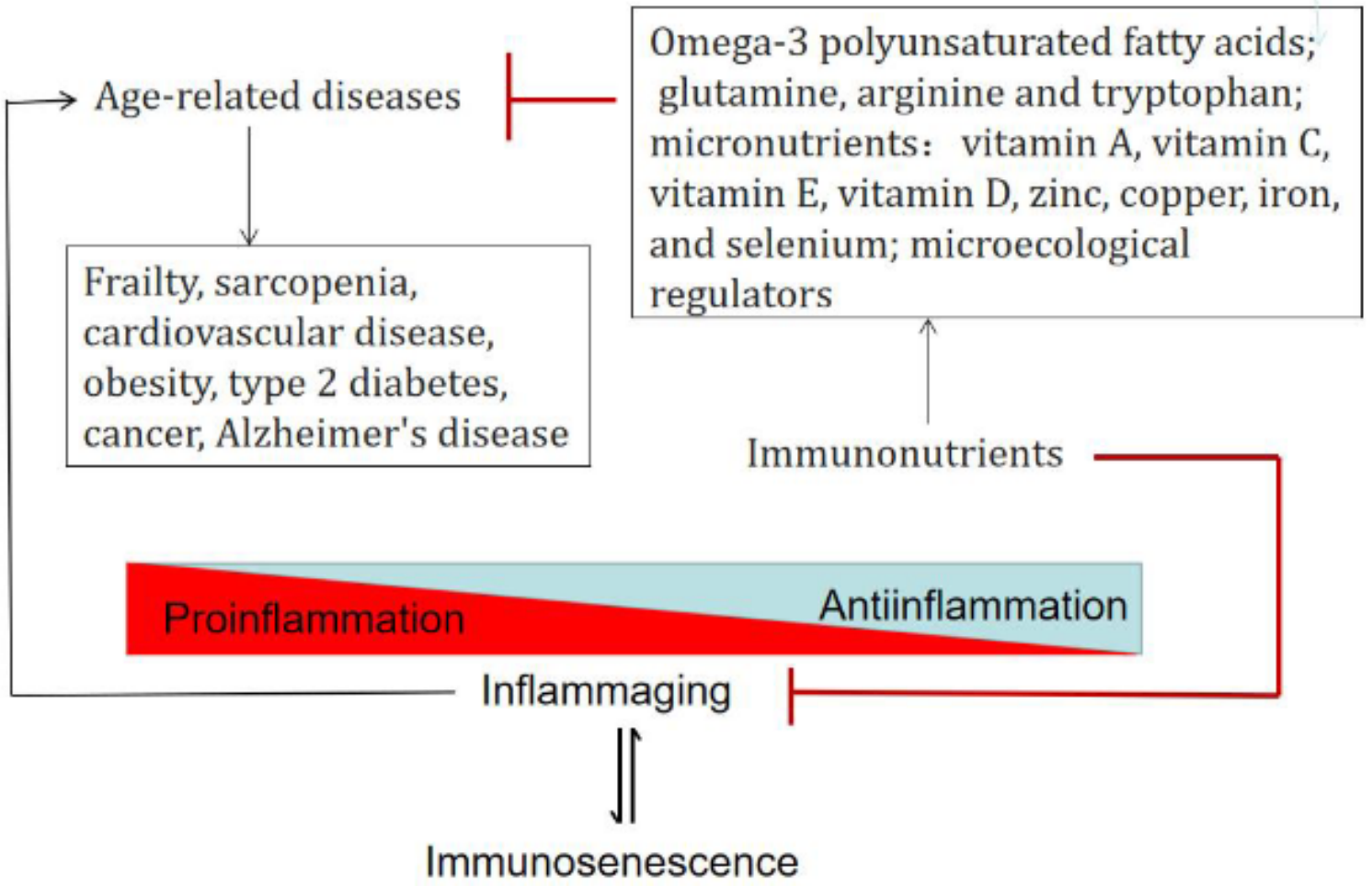
MICRONUTRIENTS

WHOLE FOODS DIET

EXERCISE

AVOIDANCE OF TOXINS

Immunonutrition



Successful Strategy

In Conclusion

- Biological aging can be managed
- The gut microbiota plays a pivotal role in overcoming trends towards frailty
- Physical activity should be a life-long habit to overcome sarcopenia
- Sarcopenia is a reflection of overall metabolic health
- A complete strategy to target the 12 hallmarks of aging should consider diet, nutrition, and exercise as foundational interventions

Thank You