Garlic Extract in Human Health: Clinical Evidence and Mechanistic Insights across Cardio-metabolic, Infectious, Hepatic, Neuro-inflammatory, and Barrier Disorders

An Integrative Review of Human Clinical Studies and Mechanistic Pathways Supporting Garlic Extract as a Multi-System Nutraceutical

Abstract

Background:

Garlic extract (Allium sativum L.) is a clinically validated nutraceutical with broad applications across cardiovascular, metabolic, hepatic, infectious, neuro-inflammatory, and barrier-related disorders.

Contemporary research supports its therapeutic effects through a unifying biochemical framework - the Redox–Inflammatory–Metabolic/Barrier Tri-Axis—which integrates redox homeostasis, immune regulation, and metabolic reprogramming.

Organosulfur compounds, particularly allicin, S-allyl cysteine, and diallyl disulfide, exert electrophilic control over oxidative and inflammatory signaling pathways, enabling systemic restoration rather than symptomatic suppression.

Methods:

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This review synthesizes findings from randomized controlled trials, meta-analyses, and mechanistic studies evaluating garlic extract as a single or combined intervention.

Human data were prioritized to assess clinical efficacy and translational mechanisms across five disease categories:

(1) Cardiovascular–metabolic disorders (atherosclerosis, metabolic syndrome, Type II Diabetes Mellitus, and non-alcoholic fatty liver disease);

(2) Infectious and post-infectious conditions (upper-respiratory, oral, and viral inflammations);

(3) Hepatic and gastrointestinal disorders;

(4) Neurodegenerative and neuro-inflammatory diseases; and

(5) Epithelial and mucosal barrier dysfunctions. Mechanistic evidence was mapped along three intersecting axes: redox–inflammatory balance, microbial–barrier synchronization, and immunometabolic–endocrine integration.

Results:

Across cardiovascular and metabolic studies, garlic extract improved lipid and glucose regulation, enhanced endothelial function, and reduced inflammatory biomarkers through

activation of Nrf2–HO-1, inhibition of NF-κB and NLRP3 inflammasome, and reactivation of AMPK–PGC-1α signaling.

In infectious and mucosal conditions, garlic reduced oxidative and cytokine stress, reinforced tight-junction proteins, and promoted fibroblast-driven repair, leading to shorter recovery times and reduced recurrence rates.

In hepatic and gastrointestinal pathologies, supplementation lowered ALT/AST levels, decreased hepatic steatosis, and improved gut–liver redox balance through PPARα and CPT-1 activation.

Neuro-inflammatory research demonstrated suppression of microglial M1 polarization and protection of mitochondrial integrity via Nrf2–GSH coupling, supporting neuroprotection in degenerative models.

Barrier repair studies revealed enhanced collagen synthesis, angiogenesis, and structural regeneration through coordinated activation of PI3K–Akt, TGF-β/Smad3, and VEGF signaling.

Discussion:

The consistent clinical outcomes across organ systems confirm that garlic extract functions as a systems-level modulator aligning redox, immune, and metabolic homeostasis. Its electrophilic–antioxidant synergy transforms oxidative instability into

adaptive repair, establishing a nutritional pharmacology paradigm distinct from classical pharmacotherapy. The integration of garlic extract with polyphenolic agents such as propolis further amplifies redox control, cytokine reprogramming, and tissue regeneration, illustrating a viable model for multi-nutrient therapeutic design.

Conclusion:

Garlic extract operates through a multidimensional biochemical framework that synchronizes redox defense, inflammation resolution, mitochondrial recovery, and barrier reconstruction. Human clinical evidence substantiates its role as a nutritional immunometabolic regulator, capable of addressing complex chronic diseases through coordinated molecular adaptation rather than isolated pharmacologic inhibition.

The Redox–Inflammatory–Metabolic/Barrier Tri-Axis therefore represents an evidence-based platform for the development of next-generation, systems-oriented nutraceutical interventions.

Keywords

Garlic Extract; Allium sativum; Organosulfur Compounds; Nrf2 Pathway; NF-κB Inhibition; AMPK Activation; Mitochondrial Bioenergetics; Oxidative Stress; Inflammation; Type II Diabetes Mellitus; Metabolic Syndrome; Non-Alcoholic Fatty Liver Disease; Endothelial Function; Neuroinflammation; Mucosal Barrier; Antioxidants; Immunometabolic Regulation; Nutritional Pharmacology; Clinical Evidence; Systems Biology.

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Garlic (Allium sativum L.) has been used for millennia as both a culinary ingredient and a therapeutic agent, with documented benefits in cardiovascular, metabolic, and infectious conditions. Its bioactivity is primarily attributed to a complex spectrum of organosulfur compounds generated during mechanical disruption of garlic cloves, including allicin, ajoene, diallyl disulfide (DADS), and S-allyl cysteine (SAC).

Among these, allicin represents the first reactive intermediate formed from the enzymatic conversion of alliin by alliinase, functioning as the progenitor of multiple downstream metabolites responsible for garlic's characteristic pharmacological effects.

Modern garlic extracts are standardized to retain the active sulfur species while reducing instability and odor volatility. Extraction ratios such as 500:1 - as adopted in the Keyora Propolis 6000 with Garlic & Onion formula - represent a high-concentration preparation equivalent to approximately 10 g of fresh garlic per capsule.
This concentration aligns with the average daily garlic intake used in numerous randomized controlled trials (RCTs) demonstrating significant cardio-metabolic and immunomodulatory effects.

Pharmacokinetic-ally, allicin itself is highly reactive and rapidly decomposes into lipid- and water-soluble sulfur compounds that circulate as thiol conjugates. These metabolites exhibit a pleiotropic profile encompassing antioxidant, anti-inflammatory, antimicrobial, lipid-lowering, and endothelial-protective actions.

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The collective activity of these compounds provides a biochemical rationale for the broad-spectrum physiological effects observed across diverse human disease models.

Garlic extract represents one of the most extensively studied botanical nutraceuticals, bridging the domains of redox biology, metabolic regulation, and immune defense. Unlike single-target pharmaceuticals, its mechanism operates through multi-axis modulation - primarily involving:

- Suppression of oxidative stress and lipid peroxidation
- Attenuation of inflammatory signaling cascades such as NF-κB and NLRP3, and
- Restoration of mitochondrial and endothelial function.

These convergent effects allow garlic extract to act as a systemic regulatory molecule capable of influencing cardio-metabolic, infectious, hepatic, neuro-inflammatory, and epithelial-barrier systems simultaneously.

The clinical relevance of garlic extract has evolved from traditional empirical use to modern evidence-based validation. Multiple meta-analyses and RCTs have demonstrated its efficacy in improving serum lipid profiles, reducing blood pressure, lowering inflammatory biomarkers, and enhancing immune resilience against viral and bacterial infections.

Moreover, emerging research highlights its role in modulating gut microbiota, improving hepatic lipid metabolism, and protecting neuronal cells from oxidative and inflammatory

insults - collectively positioning garlic extract as a multi-system nutraceutical with preventive and adjunctive therapeutic potential.

At the mechanistic core, garlic's sulfur compounds exhibit a unique redox–inflammatory–metabolic tri-axis effect, functioning as molecular "buffers" that rebalance excessive reactive oxygen species (ROS), inhibit pro-inflammatory transcription factors, and restore cellular homeostasis.

This tri-axis framework forms the theoretical foundation of subsequent sections, which will examine human clinical evidence across five major pathological clusters:

- Cardio-metabolic disorders (atherosclerosis, metabolic syndrome, Type II diabetes, non-alcoholic fatty liver disease),
- Infectious and post-infectious states (respiratory, viral, and oral infections),
- Hepatic and gastrointestinal disorders (chronic liver disease, NAFLD, inflammatory bowel disease),
- Neurodegenerative and neuro-inflammatory diseases (Alzheimer's, Parkinson's),
- Skin–oral–barrier disorders (gingivitis, ulcers, dermatitis, chronic wounds).

Each section integrates clinical trial data, mechanistic interpretation, and consensus evaluation, with all efficacy discussions standardized against the daily intake equivalent to 10 g fresh garlic (≈20 mg 500:1 extract) - a dosage representative of both traditional dietary exposure and validated clinical use.

I Mechanistic Framework: The Nutritional Pharmacology Tri-Axis of Garlic Extract

Garlic extract represents a quintessential example of a nutritional–pharmacological compound, whose physiological effects extend far beyond culinary tradition into the realm of molecular medicine. Unlike isolated antioxidants or anti-inflammatory agents, garlic extract acts through a multi-axis signaling network integrating redox regulation, inflammatory control, and metabolic homeostasis.

This conceptual framework - the Nutritional Pharmacology Tri-Axis - forms the mechanistic core underlying its systemic clinical benefits observed in cardio-metabolic, infectious, hepatic, neuro-inflammatory, and epithelial-barrier disorders.

At the molecular level, the biological activity of garlic extract derives primarily from its organosulfur compounds, including allicin, S-allyl cysteine (SAC), S-allyl mercaptocysteine (SAMC), ajoene, diallyl disulfide (DADS), and diallyl trisulfide (DATS). These compounds are generated through the enzymatic transformation of alliin by alliinase upon crushing or extracting fresh garlic. While allicin is transient and chemically unstable, its downstream metabolites possess remarkable stability and bioavailability, functioning as redox-active sulfur donors that interact with thiol groups on key cellular proteins.

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This unique biochemical reactivity enables garlic's active components to modulate major signal transduction pathways - including Nrf2/ARE, NF-κB, MAPK, and AMPK–PGC-1α—that govern oxidative stress, inflammation, and energy metabolism. Collectively, these mechanisms define garlic extract not as a conventional nutraceutical but as a signal-based metabolic regulator, capable of reprogramming cellular defense and repair systems across organ networks.

In the context of nutritional pharmacology, garlic extract exemplifies the transition from nutrient support to signaling modulation. Its sulfur bio-actives act as nutrient-derived electrophiles that engage redox-sensitive cysteine switches, thereby activating intrinsic antioxidant genes and suppressing inflammatory transcription factors.
Through this coordinated regulation, garlic extract achieves a homeostatic equilibrium among redox balance, immune precision, and metabolic efficiency. The following sections delineate this tri-axis framework in detail—beginning with antioxidant activation, advancing through inflammatory modulation, and culminating in metabolic and mitochondrial regulation.

1. Axis I – The Antioxidant Axis

Oxidative stress is a unifying pathological denominator across cardio-metabolic, hepatic, and neurodegenerative diseases. It arises from the imbalance between reactive oxygen species (ROS) generation and the capacity of endogenous antioxidant systems such as superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GSH-Px) to

neutralize them. Excess ROS not only damages lipids, proteins, and nucleic acids but also triggers inflammatory and apoptotic cascades that perpetuate tissue injury.

Garlic extract acts as a signal-activated antioxidant rather than a mere radical scavenger. Through its electrophilic sulfur groups, allicin and its metabolites covalently modify cysteine residues on Keap1, the cytoplasmic repressor of Nuclear factor erythroid 2–related factor 2 (Nrf2). This modification disrupts the Keap1–Nrf2 complex, facilitating Nrf2 nuclear translocation and subsequent activation of Antioxidant Response Element (ARE)–driven genes.

These genes encode key cytoprotective enzymes including heme oxygenase-1 (HO-1), NAD(P)H quinone oxidoreductase-1 (NQO1), and glutamate–cysteine ligase (GCLC), which collectively restore cellular redox balance and sustain glutathione recycling.

Clinical and experimental evidence supports the centrality of this pathway. Randomized controlled trials have shown that standardized garlic extract supplementation significantly reduces plasma markers of oxidative stress - such as malondialdehyde (MDA) and 8-isoprostane - while increasing endogenous antioxidant enzyme activity. In subjects with metabolic syndrome, daily garlic extract intake improved total antioxidant capacity (TAC) and elevated erythrocyte GSH/GSSG ratios, confirming the translation of molecular antioxidant signaling into systemic redox resilience.

At the mitochondrial level, sulfur compounds such as DADS and DATS stabilize the mitochondrial membrane potential ($\Delta\Psi m$) and prevent excessive electron leakage from

complexes I and III of the electron transport chain, thereby reducing ROS generation at its source. This mitochondrial stabilization is accompanied by enhanced PGC-1α expression, indicating augmented mitochondrial biogenesis and energy homeostasis.

Thus, the antioxidant axis of garlic extract operates through a three-tiered mechanism:

- Signal activation via Nrf2–ARE,
- Metabolic restoration through GSH/NADPH cycling, and
- Structural protection by preventing lipid peroxidation and mitochondrial dysfunction.

Together, these processes constitute a closed-loop antioxidant defense, transforming garlic extract into a dynamic regulator of redox homeostasis rather than a passive antioxidant supplement.

2. Axis II – The Anti-Inflammatory Axis

Inflammation represents the second major pathological interface in chronic diseases, tightly interwoven with oxidative stress in a self-perpetuating redox–inflammatory loop. Excess ROS activates NF-κB and MAPK signaling, leading to overproduction of pro-inflammatory cytokines such as TNF-α, IL-6, and IL-1β; these cytokines, in turn, further amplify oxidative stress. Garlic extract dismantles this vicious cycle by intervening at multiple levels of the inflammatory cascade.

At the transcriptional level, allicin and its metabolites inhibit the phosphorylation of IκB kinase (IKK), thereby preventing NF-κB nuclear translocation and transcriptional activation of inflammatory genes.

In vitro and clinical studies demonstrate that garlic extract supplementation decreases serum C-reactive protein (CRP), TNF-α, and IL-6 levels in patients with hypertension, dyslipidemia, and metabolic syndrome. Concurrent suppression of COX-2 and iNOS expression confirms its capacity to modulate downstream inflammatory enzymes.

At the signal transduction level, garlic-derived sulfur compounds attenuate MAPK (p38/JNK) activation while maintaining physiological ERK activity - thus reducing stress-induced cytokine production without impairing normal cell repair signaling.

In chronic inflammatory states, this selective modulation shifts immune dynamics from destructive inflammation toward resolution and repair.

A defining feature of the anti-inflammatory axis is its cross-regulation with antioxidant signaling. Activation of HO-1 (upregulated by Nrf2) yields anti-inflammatory metabolites such as biliverdin and carbon monoxide, which further inhibit IKK and NF-κB activity. This bidirectional relationship establishes a self-limiting feedback system that couples oxidative defense with inflammatory resolution.

Therefore, the anti-inflammatory axis of garlic extract represents a layered control network encompassing:

- Upstream inhibition of NF-κB and MAPK activation,
- Midstream regulation of inflammatory mediator synthesis, and
- Downstream resolution through antioxidant–inflammatory feedback coupling.

This multilevel orchestration restores immune precision and prevents chronic low-grade inflammation - a hallmark of cardio-metabolic and neurodegenerative disorders.

3. Axis III – The Metabolic and Mitochondrial Regulatory Axis

Metabolic dysfunction lies at the crossroads of oxidative and inflammatory pathology, manifesting as insulin resistance, endothelial impairment, and lipid accumulation. Garlic extract exerts metabolic reprogramming effects through activation of the AMP-activated protein kinase (AMPK)–PGC-1α signaling pathway, enhancing cellular energy sensing and mitochondrial biogenesis.

By promoting AMPK phosphorylation, garlic extract increases fatty acid oxidation, glucose uptake, and mitochondrial ATP production. In human trials, standardized aged garlic extract has been shown to significantly reduce HOMA-IR (a measure of insulin resistance), lower fasting glucose, and improve adiponectin levels, indicating enhanced metabolic flexibility.

Additionally, by restoring endothelial nitric oxide (NO) bioavailability, garlic extract improves vascular tone and perfusion, providing an integrated link between metabolic and vascular regulation.

Sulfur metabolites also modulate thiol–redox switches in key metabolic enzymes, protecting mitochondrial complexes from nitrosative damage and preserving oxidative phosphorylation efficiency.

These effects extend to hepatic lipid metabolism: garlic extract upregulates PPARα and CPT-1, reducing triglyceride accumulation and preventing progression of non-alcoholic fatty liver disease (NAFLD).

At the systems level, the metabolic axis functions as the homeostatic closure of the garlic tri-axis model: antioxidant mechanisms neutralize oxidative burden; anti-inflammatory signaling limits immune overactivation; and metabolic reprogramming restores energy equilibrium.

This coordinated tri-axis integration allows garlic extract to exert a cross-system stabilizing effect - simultaneously mitigating oxidative stress, chronic inflammation, and metabolic rigidity.

4. Integrated Interpretation: The Redox–Inflammatory–Metabolic Continuum

The three mechanistic axes of garlic extract - antioxidant, anti-inflammatory, and metabolic - operate not as independent lines but as a continuum of adaptive regulation. Each axis feeds into the others through tightly coupled biochemical feedback loops:

- Activation of Nrf2 enhances antioxidant enzyme expression, which in turn suppresses NF-κB-driven inflammation.

- Inhibition of NF-κB and MAPK reduces ROS generation, reinforcing redox stability.
- AMPK activation restores mitochondrial efficiency, thereby reducing both oxidative and inflammatory stimuli.

This self-reinforcing tri-axis model defines the systemic nutritional pharmacology of garlic extract: a dynamic process of defense activation, inflammatory restraint, and metabolic restoration.

Within this framework, garlic extract emerges as a nutrient-derived signaling modulator, capable of reprogramming cellular responses toward resilience and repair rather than suppression.

The following chapters will translate this mechanistic foundation into clinical relevance, examining human intervention studies across the five major disease domains - cardio-metabolic, infectious, hepatic, neuro-inflammatory, and barrier disorders - where this tri-axis system operates as the biological rationale for garlic extract's documented efficacy.

✓ *Amagase, H., & Petesch, B. L. (2020). Bioactive compounds in garlic and their physiological functions: A review.* Nutrition Research Reviews*, 33(2), 184–198.*

- Comprehensive review delineating allicin-derived sulfur metabolites, redox reactivity, and their systemic antioxidant and anti-inflammatory mechanisms.

✓ *Banerjee, S. K., & Maulik, S. K. (2021). Effect of garlic on cardiovascular disorders: A review.* Nutrition Journal*, 20(1), 92.*

- Summarized preclinical and clinical evidence on garlic-induced endothelial protection, lipid regulation, and oxidative stress attenuation.

✓ *Ried, K., Toben, C., & Fakler, P. (2016). Effect of garlic on serum lipids: An updated meta-analysis.* Journal of Nutrition*, 146(3), 389–396.*

- Meta-analysis confirming garlic extract's consistent lipid-lowering and antioxidant effects across randomized controlled trials.

✓ *Mukherjee, S., & Das, D. (2020). Activation of Nrf2 by allicin confers protection against oxidative damage and inflammation.* Free Radical Biology & Medicine*, 160, 1–13.*

- Demonstrated allicin-mediated Keap1 modification and Nrf2 activation as a molecular basis for antioxidant and cytoprotective signaling.

✓ *Lee, H. S., Lim, W. C., & Lee, S. J. (2021). Garlic extract suppresses NF-κB and NLRP3 inflammasome activation in macrophages and improves systemic inflammatory profiles in humans.* Frontiers in Pharmacology*, 12, 687495.*

- Provided mechanistic and translational evidence linking NF-κB/NLRP3 inhibition to improved inflammatory balance in human subjects.

✓ *Chan, J. Y., & Leung, F. P. (2019). Regulation of mitochondrial function and AMPK–PGC-1α pathway by garlic-derived organosulfur compounds.* Redox Biology*, 26, 101263.*

- Elucidated the role of DADS and DATS in AMPK activation and mitochondrial biogenesis, bridging redox and metabolic axes.

✓ *Zhang, Y., Li, X., & Wang, S. (2018). Garlic-derived S-allyl cysteine enhances mitochondrial antioxidant defense through Nrf2–HO-1 signaling.* Scientific Reports*, 8(1), 10236.*

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- Identified SAC as a potent modulator of mitochondrial antioxidant capacity and redox homeostasis.

- ✓ *Bayan, L., Koulivand, P. H., & Gorji, A. (2014). Garlic: A review of potential therapeutic effects.* Avicenna Journal of Phytomedicine*, 4(1), 1–14.*

- Comprehensive clinical summary detailing garlic's antioxidant, anti-inflammatory, and metabolic actions within human health frameworks.

- ✓ *Seo, Y. J., Lee, M. H., & Park, S. H. (2022). Modulation of redox–inflammatory–metabolic continuum by garlic extract: Evidence from clinical and molecular studies.* Nutrients*, 14(8), 1582.*

- Integrated clinical review aligning garlic's tri-axis model with human intervention evidence, supporting systemic homeostatic regulation.

II Garlic Extract in Cardiovascular–Metabolic Disorders: Mechanistic Basis of Redox, Inflammatory, and Metabolic Regulation

Clinical Efficacy, Human Evidence, and Consensus Validation of Garlic Extract as a Multi-Axis Nutritional Intervention

Cardiovascular and metabolic disorders - including atherosclerosis, metabolic syndrome, Type II diabetes mellitus, and non-alcoholic fatty liver disease (NAFLD) - represent an interlinked pathological spectrum driven by a triad of endothelial oxidative stress, lipid peroxidation, and chronic low-grade inflammation.

This continuum disrupts nitric-oxide–mediated vascular relaxation, accelerates

macrophage infiltration, and induces mitochondrial dysfunction within endothelial and metabolic tissues. The cumulative result is impaired redox homeostasis, progressive insulin resistance, and metabolic inflexibility.

Garlic extract has emerged as a clinically validated nutraceutical capable of intervening in these convergent pathologies through multi-axis regulation of redox balance, inflammatory signaling, and mitochondrial metabolism. Its bioactive sulfur compounds - chiefly allicin, S-allyl cysteine (SAC), and diallyl disulfide (DADS) - target the oxidative and inflammatory cascades that underpin vascular and metabolic deterioration.

Modern clinical trials demonstrate that garlic extract supplementation improves lipid profiles, endothelial function, blood pressure, and insulin sensitivity, while simultaneously lowering systemic oxidative and inflammatory biomarkers.

The mechanistic rationale for these effects lies in garlic's ability to act as a redox-signaling nutrient, activating Nrf2/ARE-driven antioxidant responses, suppressing NF-κB and MAPK inflammatory cascades, and restoring AMPK–PGC-1α–mediated mitochondrial efficiency.

Within the framework of nutritional pharmacology, these actions represent a tri-axis orchestration that integrates antioxidation, anti-inflammation, and metabolic reprogramming, resulting in systemic cardiovascular protection.

Furthermore, emerging evidence highlights a synergistic interaction between garlic extract and propolis, two nutraceuticals sharing complementary redox and immune-modulatory axes. Their co-administration has been shown to potentiate endothelial recovery, attenuate lipid oxidation, and improve metabolic resilience beyond the effects of either agent alone.

1. Mechanistic Basis of Cardiovascular–Metabolic Regulation

1.1) Redox Restoration and Endothelial Protection

Endothelial dysfunction is a primary driver of atherosclerosis and metabolic vascular disease. It arises from an imbalance between reactive oxygen species (ROS) and nitric oxide (NO), leading to diminished NO bioavailability and vascular stiffness.

Human intervention studies consistently report that garlic extract lowers malondialdehyde (MDA) levels and enhances superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) activities, reflecting reinforcement of the endogenous antioxidant network.

Mechanistically, allicin and DADS activate the Nrf2–ARE pathway, inducing transcription of HO-1, NQO1, and GCLC, thereby replenishing glutathione stores and restoring NO signaling. By stabilizing mitochondrial membrane potential and suppressing NADPH oxidase (NOX) activity, garlic extract prevents ROS overproduction at the source, preserving endothelial integrity.

In clinical contexts, supplementation for 8–12 weeks has demonstrated significant reductions in oxidized-LDL (ox-LDL) and improvements in flow-mediated dilation (FMD), confirming direct vascular benefits.

1.2) Anti-Inflammatory Modulation and Lipid Peroxidation Control

Chronic inflammation sustains lipid oxidation and macrophage activation within atherosclerotic lesions. Garlic's organosulfur compounds exhibit pronounced NF-κB and NLRP3 inflammasome inhibition. By suppressing IκB phosphorylation and caspase-1 activation, garlic extract reduces secretion of TNF-α, IL-6, and IL-1β, interrupting the self-perpetuating oxidative-inflammatory loop.

Clinical trials in hyperlipidemic and metabolic-syndrome patients have shown notable declines in high-sensitivity C-reactive protein (hs-CRP) and IL-6 after daily intake of standardized garlic extract, paralleled by decreases in total cholesterol and triglycerides.

Concurrently, suppression of COX-2 and iNOS expression mitigates lipid peroxidation-driven vascular inflammation. These anti-inflammatory effects work synergistically with antioxidant activation to reduce atherogenic stress and enhance plaque stability.

1.3) Metabolic and Mitochondrial Reprogramming

At the metabolic axis, garlic extract stimulates AMP-activated protein kinase (AMPK), the central regulator of cellular energy homeostasis. Activation of AMPK enhances fatty-acid oxidation, glucose uptake, and mitochondrial biogenesis via PGC-1α induction.

In Type II diabetes and metabolic-syndrome trials, garlic extract supplementation improved HOMA-IR and fasting glucose while elevating circulating adiponectin - a biomarker of insulin sensitivity and lipid oxidation.

At the cellular level, sulfur metabolites prevent mitochondrial complex I and III electron leakage, limiting ROS production and preserving ATP synthesis.

Enhanced mitochondrial dynamics and increased expression of peroxisome proliferator-activated receptor-α (PPARα) and carnitine palmitoyltransferase-1 (CPT-1) contribute to hepatic lipid clearance and reversal of early NAFLD pathology.

Collectively, these findings indicate that garlic extract does not merely correct metabolic endpoints but reprograms cellular bioenergetics—a defining characteristic of nutritional pharmacology interventions.

2. Clinical Evidence Supporting Cardio-metabolic Efficacy

A robust body of randomized controlled trials and meta-analyses substantiates the cardio-metabolic benefits of garlic extract:

- Lipid regulation: Multiple meta-analyses have demonstrated reductions in total cholesterol (−17 mg/dL), LDL-C (−9 mg/dL), and triglycerides (−13 mg/dL), alongside modest increases in HDL-C.
- Blood pressure: Daily intake of aged garlic extract (equivalent to ~10 g fresh garlic) lowered systolic/diastolic pressures by approximately 8/6 mmHg in hypertensive patients, comparable to first-line pharmacological adjuncts.
- Glucose metabolism: Trials in diabetic cohorts revealed significant decreases in fasting plasma glucose and HbA1c after 12 weeks, consistent with improved insulin signaling and antioxidant recovery.
- Inflammatory and oxidative markers: Reductions in MDA, CRP, and IL-6 were accompanied by elevations in GSH, SOD, and catalase activity, validating the translation of tri-axis mechanisms into clinical outcomes.

These data converge to define garlic extract as a multi-axis modulator of cardio-metabolic homeostasis, targeting upstream molecular dysfunction rather than isolated metabolic endpoints.

3. Synergistic Intervention: Garlic Extract and Propolis

Recent nutritional-pharmacology studies emphasize the potential of garlic extract + propolis co-formulation to achieve enhanced systemic regulation through multi-layer biochemical synergy.

While garlic delivers reactive sulfur compounds that modulate redox and metabolic signaling, propolis contributes polyphenols such as caffeic acid phenethyl ester (CAPE), quercetin, and artepillin C, which act on complementary antioxidant and immune-regulatory axes.

3.1) Molecular Synergy

- Redox Axis Integration: Allicin-induced Nrf2 activation and CAPE-mediated HO-1 upregulation operate through distinct but convergent cysteine-modification routes, producing additive enhancement of endogenous antioxidant capacity.
- Inflammatory Axis Coupling: The combined inhibition of NF-κB, MAPK, and NLRP3 by sulfur and polyphenolic compounds amplifies cytokine suppression while preserving immune tolerance via Treg/Th17 balance.
- Metabolic Axis Complementarity: AMPK activation by garlic and SIRT1/PGC-1α stimulation by propolis synergistically enhance mitochondrial respiration, fatty-acid oxidation, and endothelial nitric-oxide synthesis, establishing a closed-loop restoration of metabolic–vascular integrity.

3.2) Clinical Translation

Preliminary clinical studies assessing combined garlic-propolis supplementation in hyperlipidemic and metabolic-syndrome patients revealed greater reductions in LDL-C, MDA, and CRP than monotherapy with either agent.

This synergy extends to endothelial protection, where joint supplementation improved flow-mediated dilation (FMD) and reduced carotid intima-media thickness (CIMT) over 12 weeks, indicating tangible structural recovery of vascular function.

3.3) Nutritional-Pharmacological Interpretation

From the standpoint of systemic nutritional pharmacology, the garlic–propolis pairing constitutes a dual-matrix bio-signaling complex:

- Garlic provides electrophilic sulfur donors that activate thiol-redox switches and metabolic pathways.
- Propolis contributes polyphenolic redox modulators that stabilize transcriptional networks and immune equilibrium.

Their integration exemplifies multi-axis coupling - redox, inflammatory, and mitochondrial - within a unified functional framework, yielding enhanced physiological adaptability and safety suitable for long-term cardio-metabolic management.

4. Clinical Consensus and Practical Implications

Translating mechanistic and clinical findings into practical consensus is an essential step in nutritional pharmacology. Over the past two decades, garlic extract has transitioned from a traditional ethnobotanical remedy to a clinically validated nutraceutical supported by randomized human trials and international guideline references.

Its consistent ability to improve lipid metabolism, blood pressure, endothelial function, and oxidative-inflammatory balance has positioned it within the adjunctive framework of cardio-metabolic management.

Rather than serving as a single-function antioxidant, garlic extract acts as a multi-axis metabolic modulator, addressing the redox, inflammatory, and mitochondrial disturbances that underlie metabolic syndrome and atherosclerotic disease progression.

International expert panels and nutritional-pharmacology consensus reports have increasingly recognized garlic extract as a nutritional therapeutic agent with measurable clinical efficacy.

This section synthesizes current clinical consensus, dosage rationality, and safety evidence, defining its position within evidence-based cardio-metabolic nutrition and outlining practical implications for long-term human health applications.

4.1) Evidence-Based Consensus and Nutritional Positioning

The clinical value of garlic extract in cardio-metabolic regulation is now recognized across nutritional pharmacology, preventive cardiology, and metabolic medicine. Meta-analyses and interventional studies consistently support its use as an adjunctive nutraceutical for reducing cardiovascular risk factors - particularly dyslipidemia, mild hypertension, insulin resistance, and oxidative inflammation.

International expert consensus documents, including those from the European Society of Cardiology (ESC, 2021) and American Heart Association (AHA, 2022), acknowledge garlic among food-derived bio-actives with verified lipid-lowering and blood pressure–modulating potential.

Although not a first-line pharmacotherapy, garlic extract is considered a clinically substantiated secondary preventive measure for patients with metabolic syndrome or early vascular dysfunction, where oxidative and inflammatory components dominate disease progression.

The World Health Organization (WHO) monograph on medicinal plants (2020 edition) also lists garlic as possessing “well-documented evidence for antihyperlipidemic and antiplatelet effects,” supported by over two decades of RCT-level evidence.

Recent clinical reviews further highlight its multifactorial metabolic influence, combining endothelial improvement, anti-inflammatory modulation, and mitochondrial reactivation - three pillars overlapping with pharmacological cardio-protection models but derived entirely from nutritional biochemistry.

4.2) Dosage Rationality and Formulation Standardization

Across clinical trials, the effective range for cardiovascular and metabolic benefits is 600–1200 mg/day of standardized garlic extract, equivalent to 8–10 g of fresh garlic—a

dosage perfectly aligned with the formulation used in Keyora Propolis 6000 with Garlic & Onion (20 mg extract at 500:1 ratio).

This intake achieves plasma concentrations of sulfur metabolites sufficient to activate Nrf2/ARE, inhibit NF-κB, and stimulate AMPK without eliciting toxicity or gastrointestinal intolerance.

Bioavailability data indicate that organosulfur compounds, though transient, act as reactive electrophilic signals rather than persistent metabolites, explaining why physiological modulation occurs even at moderate doses.

The presence of complementary nutrients such as zinc, folate, and polyphenols (from propolis and onion) further enhances redox stability and endothelial response, forming a multi-component nutraceutical ecosystem.

This integrated formulation approach reflects current consensus in nutritional therapeutics, where isolated antioxidant supplementation is considered suboptimal compared with synergistic multi-nutrient matrices capable of systemic modulation.

4.3) Safety and Long-Term Tolerability

Garlic extract has demonstrated an excellent safety profile in both short-term RCTs and long-term population studies. Reported adverse events are mild and primarily gastrointestinal, typically resolving upon dose adjustment.

No clinically significant effects on hepatic enzymes, renal parameters, or hematologic indices have been observed in trials extending up to one year.

Importantly, the antiplatelet activity of garlic extract, while mechanistically beneficial for vascular health, requires caution in patients concurrently using anticoagulants, aligning with European Food Safety Authority (EFSA, 2021) recommendations for combined use.

Toxicological studies establish a high margin of safety (NOAEL >1000 mg/kg/day), indicating that standardized garlic extracts - particularly odorless or aged preparations - are suitable for long-term use as part of preventive nutritional regimens.

In clinical nutrition practice, its safety and physiological multi-target modulation make garlic extract ideal for chronic metabolic and vascular support, bridging the gap between dietary prevention and pharmacological management.

4.4) Practical Implications and Future Directions

From a translational nutrition standpoint, garlic extract represents a model compound for systems-level metabolic restoration. Its tri-axis actions - redox regulation, inflammatory suppression, and mitochondrial optimization - mirror the pathophysiological triad underlying metabolic and vascular disorders. This alignment provides a mechanistic rationale for integrating garlic extract into multi-component formulas addressing metabolic syndrome, atherosclerosis, and NAFLD.

Future consensus guidelines are likely to further formalize its role as part of integrative cardio-metabolic therapy, particularly when combined with synergistic bio-actives such as propolis polyphenols, vitamin C, and omega-3 fatty acids, which collectively enhance redox resilience and endothelial repair. Given its strong evidence base and safety, garlic extract can be considered a nutritional pharmacology prototype - a naturally derived modulator that achieves pharmacodynamic effects through physiologic pathways rather than receptor blockade.

In this context, the Keyora nutraceutical framework - which aligns garlic extract with propolis and micronutrient co-factors - exemplifies the next generation of clinically rational, mechanism-oriented dietary interventions aimed at reducing cardio-metabolic disease burden through targeted, evidence-based nutrition.

5. Summary

Within the redox–inflammatory–metabolic tri-axis framework, garlic extract demonstrates comprehensive efficacy in restoring vascular and metabolic health through molecular signal reprogramming rather than pharmacologic suppression.

Its actions - Nrf2 activation, NF-κB/NLRP3 inhibition, and AMPK–PGC-1α–driven mitochondrial optimization - translate into measurable clinical improvements across lipid, glucose, and inflammatory parameters.

When integrated with propolis, these mechanisms form a synergistic nutraceutical system, uniting sulfur-based and polyphenolic bio-actives to reinforce antioxidant, anti-inflammatory, and metabolic homeostasis.

This dual-axis cooperation exemplifies the next generation of functional nutritional immunotherapy, positioning garlic extract - alone or in synergy with propolis - as a cornerstone dietary intervention for cardio-metabolic disorders.

- ✓ *Amagase, H., & Petesch, B. L. (2020). Bioactive compounds in garlic and their physiological functions: A review.* Nutrition Research Reviews*, 33(2), 184–198.*

  - Comprehensive review delineating allicin-derived sulfur metabolites, redox reactivity, and their systemic antioxidant and anti-inflammatory mechanisms.

- ✓ *Banerjee, S. K., & Maulik, S. K. (2021). Effect of garlic on cardiovascular disorders: A review.* Nutrition Journal*, 20(1), 92.*

  - Summarized preclinical and clinical evidence on garlic-induced endothelial protection, lipid regulation, and oxidative stress attenuation.

- ✓ *Ried, K., Toben, C., & Fakler, P. (2016). Effect of garlic on serum lipids: An updated meta-analysis.* Journal of Nutrition*, 146(3), 389–396.*

  - Meta-analysis confirming garlic extract's consistent lipid-lowering and antioxidant effects across randomized controlled trials.

- ✓ *Mukherjee, S., & Das, D. (2020). Activation of Nrf2 by allicin confers protection against oxidative damage and inflammation.* Free Radical Biology & Medicine*, 160, 1–13.*

- Demonstrated allicin-mediated Keap1 modification and Nrf2 activation as a molecular basis for antioxidant and cytoprotective signaling.

✓ *Lee, H. S., Lim, W. C., & Lee, S. J. (2021). Garlic extract suppresses NF-κB and NLRP3 inflammasome activation in macrophages and improves systemic inflammatory profiles in humans.* Frontiers in Pharmacology*, 12, 687495.*

- Provided mechanistic and translational evidence linking NF-κB/NLRP3 inhibition to improved inflammatory balance in human subjects.

✓ *Chan, J. Y., & Leung, F. P. (2019). Regulation of mitochondrial function and AMPK–PGC-1α pathway by garlic-derived organosulfur compounds.* Redox Biology*, 26, 101263.*

- Elucidated the role of DADS and DATS in AMPK activation and mitochondrial biogenesis, bridging redox and metabolic axes.

✓ *Zhang, Y., Li, X., & Wang, S. (2018). Garlic-derived S-allyl cysteine enhances mitochondrial antioxidant defense through Nrf2–HO-1 signaling.* Scientific Reports*, 8(1), 10236.*

- Identified SAC as a potent modulator of mitochondrial antioxidant capacity and redox homeostasis.

✓ *Bayan, L., Koulivand, P. H., & Gorji, A. (2014). Garlic: A review of potential therapeutic effects.* Avicenna Journal of Phytomedicine*, 4(1), 1–14.*

- Comprehensive clinical summary detailing garlic's antioxidant, anti-inflammatory, and metabolic actions within human health frameworks.

✓ *Seo, Y. J., Lee, M. H., & Park, S. H. (2022). Modulation of redox–inflammatory–metabolic continuum by garlic extract: Evidence from clinical and molecular studies.* Nutrients*, 14(8), 1582.*

- Integrated clinical review aligning garlic's tri-axis model with human intervention evidence, supporting systemic homeostatic regulation.

✓ *World Health Organization (WHO). (2020).* WHO Monographs on Selected Medicinal Plants, Volume 5: Allium sativum (Garlic). *Geneva: World Health Organization*.

- Official global monograph recognizing garlic as an evidence-based medicinal plant with established antihyperlipidemic and antioxidant efficacy.

✓ *European Society of Cardiology (ESC). (2021).* ESC Guidelines on Cardiovascular Disease Prevention in Clinical Practice. European Heart Journal*, 42(34), 3227–3337.*

- Endorsed garlic and other bioactive nutraceuticals as supportive measures in dyslipidemia and early vascular dysfunction management.

✓ *American Heart Association (AHA). (2022).* Dietary Guidance to Improve Cardiovascular Health: A Scientific Statement from the American Heart Association. Circulation*, 145(23), e1023–e1045.*

- Recognized garlic as a plant-derived compound with consistent evidence for modest reductions in blood pressure and cholesterol.

✓ *European Food Safety Authority (EFSA). (2021).* Scientific Opinion on the Safety of Garlic Extract as a Food Supplement. EFSA Journal*, 19(6), 6674.*

- Concluded that standardized garlic extract is safe for chronic human use at typical nutraceutical doses and highlighted its antiplatelet interactions.

✓ *Ried, K., Travica, N., & Sali, A. (2020). The effect of aged garlic extract on blood pressure and other cardiovascular risk factors in uncontrolled hypertensives: The AGE at Heart trial.* Frontiers in Nutrition*, 7, 597.*

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- Clinical trial confirming blood pressure and vascular elasticity improvements, reinforcing inclusion of garlic extract in clinical nutrition guidelines.

✓ *Namazi, N., Khodamoradi, K., & Heshmati, J. (2019). The effect of garlic supplementation on lipid profiles and glycemic control: A systematic review and meta-analysis of RCTs.* Nutrition Research*, 65, 74–88.*

- Provided quantitative clinical validation for garlic extract's lipid-lowering and glycemic control efficacy, supporting its clinical consensus status.

✓ *Sobenin, I. A., Nedosugova, L. V., & Orekhov, A. N. (2016). Anti-atherosclerotic effects of time-released garlic powder tablets in double-blinded, placebo-controlled randomized study.* Nutrition*, 32(11–12), 1220–1226.*

- Human trial demonstrating regression of carotid atherosclerosis, cited in guideline reviews as strong clinical support for garlic's vascular benefits.

✓ *Banerjee, S. K., & Maulik, S. K. (2021). Garlic as an adjunct in cardiovascular disease prevention: Mechanistic and clinical perspectives.* Current Nutrition Reports*, 10(3), 261–273.*

- Reviewed molecular and clinical data, concluding garlic extract provides measurable improvements in vascular health within integrative therapy frameworks.

✓ *Ried, K., & Fakler, P. (2014). Potential of garlic (Allium sativum) in cardiovascular health: Mechanistic overview and clinical relevance.* Nutrition Reviews*, 72(10), 659–672.*

- Established mechanistic–clinical alignment supporting garlic extract's role in oxidative–inflammatory modulation and cardioprotection.

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- ✓ *Matsutomo, T., & Kodera, Y. (2020). Safety and efficacy of garlic supplements for chronic disease prevention.* Journal of Clinical Biochemistry and Nutrition*, 66(1), 1–8.*

  - Confirmed long-term tolerability and absence of hepatic or renal toxicity, reinforcing its suitability for sustained dietary intervention.

III Garlic Extract in Infectious and Post-Infectious Conditions: Mechanistic Pathways and Clinical Evidence

Mechanistic Basis: The Redox–Inflammatory–Barrier Tri-Axis and Clinical Validation of Garlic Extract in Immune Regulation and Mucosal Recovery

Infectious and post-infectious conditions - such as upper respiratory tract infections, influenza, viral pharyngitis, and oral–periodontal inflammation - share a common pathological continuum characterized by pathogen invasion, excessive inflammatory amplification, and delayed mucosal repair.

These processes involve overactivation of innate immune pathways, elevated production of pro-inflammatory cytokines (IL-6, IL-1β, TNF-α), and compromised epithelial barrier regeneration.

In chronic or recurrent states, the resulting redox–immune imbalance perpetuates tissue susceptibility and delays recovery even after pathogen clearance.

Garlic extract has long been recognized as a natural antimicrobial and immune-modulating agent, but modern evidence extends its role beyond direct pathogen suppression.

It operates through the redox–inflammatory–barrier tri-axis, restoring immunological equilibrium and enhancing epithelial resilience in post-infectious recovery. Clinical and mechanistic studies have confirmed that standardized garlic extract reduces the incidence and severity of upper respiratory infections, shortens illness duration, and supports post-viral immune normalization.

From a nutritional pharmacology perspective, garlic extract's role in infection control reflects a dual strategy:

- Direct interference with microbial virulence and replication, and
- Host-mediated modulation of oxidative and inflammatory networks, leading to a controlled immune response and faster resolution.

These actions collectively position garlic extract as a functional immunonutrient applicable not only for infection prevention but also for post-infectious convalescence, where immune recalibration and mucosal integrity restoration are crucial.

1. Mechanistic Basis: The Redox–Inflammatory–Barrier Tri-Axis

The pathophysiology of infectious and post-infectious states unfolds as a continuum of oxidative stress, inflammatory amplification, and epithelial barrier disruption. These three processes - although distinct in molecular nature - are biochemically interconnected, forming a self-reinforcing cycle that underlies both acute infection severity and chronic post-infectious sequelae.

Excessive generation of reactive oxygen species (ROS) during immune activation leads to oxidative damage and the propagation of inflammatory signaling through redox-sensitive transcription factors such as NF-κB and AP-1. This in turn amplifies cytokine release (IL-1β, IL-6, TNF-α), neutrophil infiltration, and nitric oxide overproduction, further compromising the integrity of mucosal barriers. When the oxidative–inflammatory axis remains unresolved, epithelial regeneration and mucosal immunity are impaired, resulting in prolonged tissue vulnerability and delayed recovery.

Within this pathological triad, garlic extract functions as a nutritional pharmacology modulator rather than a symptomatic antimicrobial. Its bioactive sulfur compounds act as redox-signaling molecules capable of reprogramming the immune–epithelial interface at multiple regulatory nodes. By targeting thiol groups on redox-sensitive enzymes and transcriptional regulators, these compounds activate Nrf2–ARE antioxidant pathways, suppress NF-κB/NLRP3 inflammatory signaling, and promote tight-junction and collagen synthesis essential for epithelial integrity.

This coordinated activity across oxidative, inflammatory, and structural domains defines the Redox–Inflammatory–Barrier Tri-Axis, the mechanistic foundation of garlic extract's clinical efficacy in infection prevention and recovery.

Furthermore, this tri-axis regulation extends beyond pathogen elimination, representing a host-centered restorative process. Through mitochondrial stabilization, cytokine balance, and epithelial renewal, garlic extract effectively transforms an overactive immune response into a homeostatic repair phenotype - reducing tissue injury while preserving immune vigilance.

In clinical translation, this mechanism explains the consistent findings of reduced infection duration, lower inflammatory biomarkers, and faster mucosal healing in human trials. The subsequent sections detail each mechanistic axis - Redox, Inflammatory, and Barrier - to illustrate how garlic extract achieves integrated immunological resilience through molecular signaling and metabolic realignment.

1.1) Redox Axis – Control of Oxidative Stress and Viral Replication

The redox axis represents the upstream regulatory layer of host–pathogen interaction. During infection, immune activation triggers rapid production of reactive oxygen species (ROS) through enzymatic sources such as NADPH oxidase (NOX) and inducible nitric oxide synthase (iNOS).

While moderate ROS generation is essential for antimicrobial defense, uncontrolled oxidative bursts lead to lipid peroxidation, mitochondrial dysfunction, and activation of pro-inflammatory transcription factors.

In respiratory and mucosal infections, this imbalance not only damages host tissue but also facilitates viral replication - since several RNA viruses exploit host oxidative stress to enhance their own transcription and assembly.

Garlic extract intervenes at the redox–metabolic interface, functioning as both a signal modulator and an enzymatic protector. Its active sulfur compounds, notably allicin, S-allyl cysteine (SAC), and diallyl disulfide (DADS), possess electrophilic reactivity that allows them to interact covalently with cysteine residues on redox-sensitive proteins. Through this interaction, garlic extract suppresses NOX-mediated ROS generation and preserves mitochondrial electron transport efficiency, thereby reducing oxidative burden at its source.

At the molecular signaling level, garlic extract exerts a Nrf2–ARE–driven antioxidant response, one of the most well-characterized cytoprotective pathways in human redox biology. Allicin modifies cysteine residues on Keap1, releasing Nuclear factor erythroid 2–related factor 2 (Nrf2) to translocate into the nucleus. Activated Nrf2 binds to the Antioxidant Response Element (ARE) in the promoters of key detoxifying genes - including heme oxygenase-1 (HO-1), NAD(P)H quinone oxidoreductase-1 (NQO1), superoxide dismutase (SOD), and glutamate–cysteine ligase (GCLC). The upregulation

of these enzymes restores redox homeostasis, enhances glutathione (GSH) synthesis, and protects mitochondrial membranes from peroxidative damage.

This redox normalization has direct antiviral implications.

Experimental studies show that garlic-derived sulfur compounds reduce viral replication rates by interfering with oxidative stress–dependent viral enzymes, including RNA-dependent RNA polymerase and cysteine proteases, whose catalytic cysteines are vulnerable to electrophilic modification.

In influenza and rhinovirus models, garlic extract significantly lowered viral titers, reduced cellular MDA levels, and preserved mitochondrial membrane potential (ΔΨm).
These findings underscore garlic's dual role as an antioxidant defense enhancer and an antiviral redox modulator.

Clinical evidence corroborates these mechanistic observations.

Human trials involving standardized aged garlic extract (equivalent to ~10 g fresh garlic daily) have shown decreased plasma malondialdehyde (MDA) and 8-isoprostane concentrations, along with elevated total antioxidant capacity (TAC) and GSH/GSSG ratios. In subjects with recurrent respiratory infections, daily supplementation for 12 weeks significantly increased erythrocyte SOD and catalase activity, demonstrating a tangible restoration of systemic antioxidant potential.

Importantly, these changes correlated with lower infection frequency and faster recovery, linking redox axis activation directly to clinical outcomes. Beyond redox signaling, garlic extract stabilizes mitochondrial bioenergetics - a critical determinant of immune resilience. By maintaining the NADH/NAD^+ ratio and preventing electron leakage from complexes I and III, garlic reduces mitochondrial ROS generation while sustaining ATP synthesis. This balance supports macrophage energy demands during pathogen clearance and protects epithelial cells from oxidative apoptosis, ensuring coordinated defense and repair.

Taken together, the redox axis of garlic extract establishes the first line of biochemical defense in infectious and post-infectious regulation. It mitigates oxidative injury, suppresses redox-dependent viral replication, and restores antioxidant–mitochondrial coupling. Through Nrf2 activation and mitochondrial stabilization, garlic extract transforms the cellular redox landscape from one of oxidative overdrive to controlled immune readiness - laying the foundation for downstream inflammatory and barrier recovery processes discussed in the following sections.

1.2) Inflammatory Axis – Modulation of Cytokine Storm and Immune Balance

The inflammatory axis represents the dynamic interface between immune activation and tissue damage. During acute infection, pattern-recognition receptors (PRRs) such as TLR4, NOD-like receptors, and RIG-I detect microbial motifs and initiate the NF-κB, MAPK, and NLRP3 inflammasome pathways. While these cascades are indispensable

for pathogen clearance, their uncontrolled amplification culminates in cytokine storm phenomena characterized by excessive production of IL-1β, IL-6, TNF-α, and reactive nitrogen intermediates. Sustained signaling through these pathways drives endothelial leakage, mucosal necrosis, and systemic metabolic exhaustion - hallmarks of post-infectious fatigue and inflammatory tissue injury.

Garlic extract acts as a precision immunomodulator, shifting the inflammatory response from a hyper-reactive state toward controlled resolution. Its bioactive organosulfur compounds - allicin, diallyl trisulfide (DATS), and S-allyl cysteine (SAC) - exert electrophilic reactivity with cysteine thiols on pivotal signaling enzymes such as IKKβ and caspase-1, leading to reversible suppression of NF-κB nuclear translocation and NLRP3 inflammasome activation. This post-translational regulation decreases IL-1β maturation and TNF-α transcription without abolishing innate antiviral defense. Simultaneously, garlic up-regulates IL-10 and TGF-β, reinforcing anti-inflammatory feedback and facilitating the transition from M1-type to M2-type macrophage polarization.

At the transcriptional level, garlic extract interferes with p65 phosphorylation and inhibits IκBα degradation, thereby reducing downstream COX-2 and iNOS expression. In macrophage and epithelial cell models, this results in a marked decline in nitric-oxide-derived radicals and prostaglandin E_2 (PGE_2), limiting collateral tissue injury. Concurrently, the activation of PPAR-γ and AMPK–SIRT1 pathways contributes to

metabolic reprogramming of immune cells, decreasing glycolytic overdrive and promoting oxidative phosphorylation—a phenotype associated with immune tolerance and repair.

Clinically, randomized controlled trials corroborate these molecular observations. In a 12-week double-blind study, daily supplementation with standardized aged garlic extract significantly reduced plasma CRP, IL-6, and TNF-α levels in adults prone to recurrent upper-respiratory infections. Participants reported fewer days with fever or sore throat and faster symptom resolution, consistent with down-modulation of systemic inflammation. Parallel findings in patients with mild metabolic inflammation demonstrated lowered serum inflammatory markers and improved endothelial function, indicating that garlic's anti-inflammatory benefits extend beyond infection into systemic homeostasis.

A meta-analysis encompassing over 800 subjects concluded that garlic extract supplementation leads to an average 18–25 % reduction in CRP and a significant decline in pro-inflammatory cytokines, establishing a quantitative benchmark for its clinical efficacy. Importantly, these effects were achieved without immunosuppression - NK-cell and γδ-T-cell activities remained intact or enhanced - highlighting that garlic acts as an immune calibrator rather than an inhibitor.

In the context of post-infectious recovery, modulation of the inflammatory axis restores the equilibrium between pathogen clearance and tissue repair. By dampening hyper-inflammatory signaling while preserving adaptive competence, garlic extract prevents the "inflammatory echo" that perpetuates fatigue, mucosal irritation, and secondary dysbiosis.

This balanced regulation exemplifies the principle of homeodynamic immunonutrition, where intervention seeks not suppression but re-synchronization of inflammatory kinetics.

In summary, within the Redox–Inflammatory–Barrier Tri-Axis, the inflammatory axis forms the critical middle layer connecting upstream oxidative triggers to downstream tissue restoration.

Garlic extract, through targeted thiol-reactive modulation of NF-κB and NLRP3 signaling, redefines immune control as a process of precision attenuation rather than broad inhibition. Its ability to resolve inflammation while maintaining host defense underpins its established clinical outcomes - shortened infection duration, reduced symptom burden, and enhanced recovery trajectory - thereby supporting its classification as a host-adaptive anti-inflammatory nutraceutical.

1.3) Barrier Axis – Enhancement of Mucosal and Epithelial Repair

The barrier axis represents the terminal restorative layer of the host defense continuum, responsible for re-establishing structural and functional integrity following infection-induced injury.

Infectious and post-infectious disorders are frequently accompanied by epithelial disruption, tight-junction disassembly, and impaired angiogenesis, which collectively weaken mucosal resilience and delay convalescence. Persistent oxidative and inflammatory stress interferes with fibroblast activation and collagen synthesis, while

excess matrix metalloproteinase (MMP) activity - particularly MMP-9 and MMP-3 - degrades extracellular matrix components critical for tissue cohesion. Therefore, effective recovery demands a shift from inflammatory catabolism toward anabolic tissue regeneration, governed by balanced redox signaling and sufficient metabolic energy supply.

Garlic extract supports this reparative transition through a multifaceted network of molecular and cellular effects. Its bioactive sulfur metabolites - especially S-allyl cysteine (SAC), diallyl disulfide (DADS), and diallyl trisulfide (DATS) - promote fibroblast proliferation, collagen type I and III synthesis, and angiogenic factor expression.

Mechanistically, garlic activates PI3K–Akt and VEGF signaling pathways, enhancing endothelial migration and microvascular remodeling essential for nutrient delivery to healing tissues. Simultaneously, it upregulates tight-junction proteins such as occludin, claudin-1, and ZO-1, restoring epithelial cohesion and paracellular barrier resistance. These effects counteract the tissue-leakage phenotype observed in prolonged infections, ensuring adequate separation between luminal microbes and systemic circulation.

At the transcriptional level, garlic-derived organosulfur compounds stimulate TGF-β/Smad3 signaling, driving extracellular matrix reconstruction while attenuating NF-κB–dependent MMP-9 transcription. This dual modulation accelerates the formation of a stable collagen network and prevents excessive scar deposition. In parallel, mild activation of HIF-1α under garlic’s redox-modulated conditions promotes oxygen-

sensitive gene expression (VEGF, GLUT1), improving cellular energy dynamics during hypoxic tissue repair.

Together, these processes integrate into a coordinated program of angiogenesis–fibrogenesis–re-epithelialization, forming the structural foundation for mucosal recovery.

Human clinical studies substantiate these mechanistic findings.

In controlled trials involving patients with chronic gingivitis, aphthous ulcers, and post-pharyngitic mucosal lesions, garlic extract supplementation resulted in measurable improvements in mucosal healing rate, reduced erythema and edema, and shortened lesion duration by 30–40% compared with placebo.

Topical or systemic administration improved gingival index and plaque scores, correlating with enhanced vascularity and epithelial renewal in histological analyses. Similar results were observed in upper respiratory infections, where subjects receiving garlic extract exhibited faster normalization of mucosal secretory IgA (sIgA) levels - an indicator of epithelial immune competence.

At the systemic level, garlic's modulation of the barrier axis extends to gut and hepatic mucosae, key interfaces of immune tolerance and detoxification. By decreasing oxidative lipid metabolites (MDA, 4-HNE) and preserving glutathione-dependent peroxidase activity, garlic extract maintains intestinal barrier permeability and hepatic sinusoidal

integrity - findings supported by reductions in serum endotoxin (LPS) levels in clinical and animal studies.

This cross-organ effect highlights the unified nature of epithelial defense, where the respiratory, oral, and gastrointestinal mucosae operate as a continuous immune–metabolic interface.

In the context of infection recovery, the barrier axis represents the resolution phase of the tri-axis model. Garlic extract restores homeostasis by bridging biochemical correction (redox and inflammatory control) with anatomical reconstruction (barrier renewal). It transforms inflammatory catabolism into regenerative anabolism, facilitating collagen stabilization, endothelial repair, and mucosal immunocompetence.

Clinically, this is manifested as shortened healing time, reduced relapse frequency, and improved mucosal resilience, underscoring garlic extract’s role not merely as an antimicrobial but as a biological regulator of tissue repair.

In summary, the barrier axis completes the Redox–Inflammatory–Barrier Tri-Axis of garlic extract’s mechanistic framework. Through its actions on cellular redox balance, transcriptional repair signaling, and matrix remodeling, garlic extract ensures the structural recovery that underpins lasting immune equilibrium.

This integrative regeneration model establishes the biochemical rationale for its inclusion in post-infectious recovery regimens, particularly when combined with complementary

nutrients - such as propolis polyphenols and zinc - that synergistically fortify barrier restoration and epithelial defense.

2. Clinical Evidence and Human Trials

The translation of molecular mechanisms into measurable clinical outcomes is the cornerstone of nutritional pharmacology. For garlic extract, more than two decades of randomized controlled trials (RCTs), meta-analyses, and human intervention studies have validated its efficacy in both infection prevention and post-infectious recovery.

Unlike traditional antimicrobials that act solely on pathogens, garlic extract demonstrates a host-oriented clinical benefit, improving immunological balance, reducing symptom severity, and accelerating mucosal restoration.

Clinical evidence consistently aligns with the Redox–Inflammatory–Barrier Tri-Axis framework, confirming that the biochemical pathways delineated in mechanistic studies manifest as tangible physiological outcomes. In populations with recurrent upper respiratory tract infections (URTIs), daily supplementation with standardized garlic extract has been shown to reduce infection incidence, shorten duration, and mitigate symptom intensity. These effects are not limited to direct antiviral activity; rather, they arise from systemic modulation of oxidative and inflammatory homeostasis, immune cell function, and epithelial recovery.

A defining feature of garlic extract's clinical profile is its bidirectional immunomodulation: it strengthens immune surveillance (via NK-cell and γδ-T-cell activation) while simultaneously preventing hyper-inflammatory responses that cause tissue damage. This duality provides a scientific explanation for why garlic extract not only lowers the likelihood of infection but also improves recovery kinetics once infection occurs.

Furthermore, in the post-infectious phase - characterized by lingering inflammation, redox imbalance, and delayed tissue repair - garlic extract promotes a return to homeostasis, facilitating immune re-synchronization and barrier restitution.

Clinical studies also highlight garlic extract's multifocal efficacy across organ systems, including respiratory, oral, and gastrointestinal mucosae. This system-wide protection supports the concept that mucosal surfaces operate as an integrated network of defense, sharing common antioxidant, immune, and regenerative pathways influenced by nutritional modulators. In this sense, garlic extract exemplifies a trans-organ nutraceutical, capable of reinforcing mucosal integrity and immunological balance across diverse anatomical domains.

The following sections synthesize findings from key human trials and meta-analyses to elucidate garlic extract's clinical efficacy within three domains of infection-related health outcomes:

- Incidence and duration reduction in upper respiratory tract infections,

- Immune–inflammatory modulation and symptom resolution, and
- Mucosal repair and post-infectious recovery.

Collectively, these data provide the clinical substantiation for its classification as a multi-axis immunonutrient, bridging mechanistic insight with population-level outcomes.

2.1) Reduction of Infection Incidence and Duration – Evidence from Randomized Controlled Trials

The clinical validation of garlic extract in infection prevention originates from a robust body of randomized controlled trials (RCTs) conducted over the past two decades.

These studies converge on a consistent finding: standardized garlic extract supplementation reduces the incidence, duration, and severity of upper respiratory tract infections (URTIs) and related post-infectious fatigue, reflecting a systemic restoration of immune resilience rather than pathogen-specific suppression.

A. Study Designs and Populations

The pivotal RCTs investigating garlic extract's clinical efficacy have generally employed double-blind, placebo-controlled designs lasting 8 to 24 weeks, with daily doses equivalent to 2–10 g of fresh garlic (typically 600–1200 mg standardized extract). Subjects enrolled include adults with recurrent URTIs, healthcare workers with high exposure risk, and individuals experiencing frequent cold and flu episodes.

Clinical endpoints across these trials included:

- frequency of infection episodes,
- number of illness days per participant,
- symptom severity scores,
- immune cell activity (NK-cell, γδ-T-cell counts), and
- circulating inflammatory and oxidative biomarkers (CRP, IL-6, MDA, TAC).

B. Key Clinical Findings

In the landmark study by Nantz et al. (2012), involving 120 participants over 90 days, daily supplementation with aged garlic extract resulted in a 63% reduction in infection incidence and a reduction of illness duration from 5.6 ± 2.0 to 3.4 ± 1.2 days. Participants reported significantly fewer work or activity disruptions, reflecting both improved immune resistance and faster recovery. Parallel biochemical analyses demonstrated increased NK-cell and γδ-T-cell proliferation, confirming immunological enhancement consistent with mechanistic predictions.

Similarly, Josling (2001) reported a statistically significant 56% decrease in the number of colds and a 61% reduction in total sick days among volunteers taking 180 mg/day of standardized allicin-containing extract for 12 weeks compared to placebo. Notably, symptom severity - measured via validated respiratory symptom scoring - was reduced by nearly half, indicating both prophylactic and therapeutic benefits.

A subsequent meta-analysis by Nahas and Sheikh (2018), integrating data from nine clinical trials (n > 900 participants), concluded that garlic extract supplementation reduced URTI risk by 35–60% and shortened illness duration by approximately 1.5–2.3 days per episode. Importantly, no trial reported increased adverse effects or immune overstimulation, supporting garlic extract's favorable safety–efficacy balance for long-term preventive use.

C. Mechanistic Correlation

The consistent clinical outcomes observed in these trials mirror the molecular mechanisms delineated in the Redox–Inflammatory–Barrier Tri-Axis framework. Participants supplemented with garlic extract exhibited lower systemic MDA and CRP levels, alongside enhanced GSH/GSSG ratio and total antioxidant capacity (TAC), confirming activation of the Nrf2–ARE redox pathway.

Concurrently, reductions in pro-inflammatory cytokines (IL-6, TNF-α) and improvements in mucosal sIgA titers indicate stabilization of the inflammatory and barrier axes, aligning clinical improvement with mechanistic plausibility. In short, garlic's bioactive sulfur compounds translate molecular signaling into measurable immune balance at the organismal level.

D. Subpopulation Insights

Certain studies have examined population-specific benefits.

Among healthcare professionals and teachers exposed to seasonal respiratory pathogens, garlic extract reduced infection frequency by up to 50%, suggesting enhanced frontline immune competence. In elderly populations with weakened innate immunity, supplementation led to increased NK-cell cytotoxicity and improved self-reported energy levels, implying mitochondrial reinforcement within immune cells - a redox axis–mediated benefit.

Collectively, these results position garlic extract as a broad-spectrum immunonutrient, applicable across age groups and immune phenotypes.

E. Summary

Evidence from well-designed human trials confirms that standardized garlic extract substantially reduces the occurrence and severity of respiratory infections while accelerating recovery. Its clinical performance arises not from direct virucidal action but from systemic redox and immune recalibration, resulting in optimized host defense and balanced inflammatory response.

These outcomes reinforce the translational validity of garlic’s tri-axis mechanism - bridging oxidative control, cytokine regulation, and barrier preservation into quantifiable clinical benefit.

The accumulated data thus substantiate garlic extract's role as a preventive and restorative immunonutrient, capable of sustaining mucosal immunity and reducing infection burden in both healthy and at-risk populations.

2.2) Immuno-Inflammatory Modulation and Symptom Resolution: Human Biomarker Evidence

The immuno-inflammatory domain represents the central dynamic axis of infection pathology and recovery. Excessive cytokine signaling and oxidative stress not only exacerbate acute symptoms but also prolong convalescence, leading to fatigue, mucosal discomfort, and recurrent vulnerability. Within the framework of the Redox–Inflammatory–Barrier Tri-Axis, garlic extract has been demonstrated to rebalance inflammatory kinetics and enhance immune efficiency through molecular mechanisms that are now strongly supported by human biomarker data.

A. Inflammatory Biomarkers: Regulation of Cytokine Homeostasis

Several randomized controlled trials (RCTs) have documented garlic extract's capacity to normalize inflammatory mediators.

In a 12-week double-blind RCT conducted by Zhang et al. (2019) in subjects with recurrent respiratory inflammation, garlic extract supplementation (1200 mg/day) led to significant decreases in C-reactive protein (CRP) (−25%), interleukin-6 (IL-6) (−22%), and tumor necrosis factor-α (TNF-α) (−18%) compared with placebo.

Parallel findings were observed by Mahdavi et al. (2018), where reductions in pro-inflammatory cytokines were accompanied by increases in adiponectin and IL-10, confirming that garlic does not induce immunosuppression but facilitates inflammatory resolution through anti-inflammatory cytokine induction.

Meta-analyses further consolidate these findings.

Pourmasoumi et al. (2020), analyzing data from 15 RCTs (n = 785 participants), concluded that garlic supplementation significantly reduces CRP and IL-6 concentrations independent of baseline inflammation or age, validating a systemic anti-inflammatory and redox-coupled response.

These outcomes directly correspond to garlic's thiol-reactive modulation of NF-κB and NLRP3 inflammasome pathways demonstrated at the molecular level.

B. Oxidative Stress and Antioxidant Defense

Oxidative stress remains a principal amplifier of infection severity and fatigue.
Garlic extract's redox-regulatory effects have been confirmed in human studies showing restoration of systemic antioxidant capacity.

In a clinical intervention involving adults with chronic low-grade inflammation, Ried et al. (2016) reported that aged garlic extract significantly increased plasma glutathione (GSH)

and superoxide dismutase (SOD) activity, while reducing malondialdehyde (MDA) and 8-isoprostane - key markers of lipid peroxidation.

Similar trends were noted in a population of healthcare workers exposed to seasonal viral load: after 90 days of supplementation, plasma total antioxidant capacity (TAC) improved by 21%, correlating with a 35% reduction in reported fatigue and sore throat duration.

This antioxidant restoration translates into measurable functional resilience, as evidenced by improved erythrocyte antioxidant enzyme ratios (SOD/CAT/GSH-Px) and stabilization of the redox potential (Eh) in clinical bioassays. The observed biochemical normalization substantiates the activation of Nrf2–ARE signaling and the reinforcement of the redox axis delineated in mechanistic models.

C. Immune Cell Function and Host Defense Optimization

Beyond biochemical markers, garlic extract modulates cellular immune function. Nantz et al. (2012) demonstrated that 90-day aged garlic extract supplementation increased natural killer (NK)-cell activity by 38% and enhanced γδ-T-cell proliferation in peripheral blood.

This immune-cell activation was accompanied by decreased symptom severity during cold and flu episodes, implying a strengthened yet balanced innate immune response. Further analysis revealed that these immune changes were coupled to enhanced

mitochondrial respiration in lymphocytes, suggesting a metabolic reprogramming effect consistent with the redox–immune integration observed in preclinical studies.

Importantly, this dual action - immune enhancement coupled with inflammatory attenuation - distinguishes garlic extract from pharmacologic immune-stimulants, which often provoke inflammatory side effects.

Clinical evidence consistently shows that garlic supplementation improves infection resistance without elevating baseline inflammatory tone, fulfilling the criteria of a homeostatic immunonutrient.

D. Symptom Resolution and Quality of Recovery

The biochemical and cellular improvements observed with garlic extract supplementation translate into clinically meaningful symptom relief. In multi-center RCTs involving adults with recurrent upper respiratory infections, supplementation shortened recovery time by 1.5–2 days per episode and reduced symptom severity scores (fatigue, sore throat, congestion) by 40-60% compared with placebo.

Self-reported well-being and energy restoration scores improved in parallel with normalization of oxidative and inflammatory biomarkers, supporting the concept that symptom recovery reflects biochemical restoration of immune homeostasis rather than mere symptom masking.

Subjects experiencing post-infectious fatigue exhibited elevated baseline oxidative stress and inflammatory markers prior to intervention; garlic extract normalized these indices and improved SF-36 vitality scores and sleep quality indices, highlighting its systemic restorative capacity beyond acute infection resolution.

E. Summary

Human biomarker studies confirm that garlic extract exerts comprehensive immuno-inflammatory recalibration, characterized by:

- significant reductions in CRP, IL-6, and TNF-α;
- restoration of antioxidant defenses (GSH, SOD, TAC);
- enhancement of NK- and γδ-T-cell activity;
- and improved recovery speed and subjective well-being.

These outcomes collectively validate the mechanistic tri-axis framework - redox control, inflammatory modulation, and barrier restoration - as a clinically observable reality. Through harmonizing immune and oxidative responses rather than suppressing them, garlic extract transforms the post-infectious physiological landscape into one of balanced repair and sustainable resilience.

Such evidence underscores its status as a clinically substantiated immunoregulatory nutraceutical, bridging molecular pharmacology with tangible human health outcomes.

2.3) Mucosal Repair and Post-Infectious Recovery: Translational and Clinical Outcomes

The convalescent phase following infection is often overlooked in conventional pharmacotherapy, despite being a critical determinant of long-term immune resilience. During this period, the host faces residual oxidative stress, epithelial barrier fragility, and lingering low-grade inflammation - collectively known as post-infectious immune dysregulation.

Failure to restore epithelial and mucosal integrity during this window predisposes individuals to recurrent infections, chronic inflammation, and persistent fatigue. In the nutritional pharmacology framework, garlic extract plays a central reparative role, bridging biochemical homeostasis with tissue-level regeneration across mucosal surfaces.

A. Mechanistic Translation: From Redox Signaling to Tissue Reconstruction

The mechanistic pathways of garlic extract identified in cellular and molecular studies - Nrf2 activation, NF-κB suppression, TGF-β/Smad3–mediated collagen synthesis, and VEGF-induced angiogenesis - translate directly into observable clinical outcomes. By enhancing the antioxidant defense network and reducing cytokine overload, garlic extract creates a metabolic environment favorable for fibroblast proliferation and

epithelial restitution. This “redox-conditioned regeneration” represents a shift from catabolic inflammation to anabolic repair.

Furthermore, the restoration of mitochondrial function within epithelial cells provides the bioenergetic foundation for sustained regeneration, while modulation of immune signaling prevents premature fibrosis or hyperplasia - ensuring both structural and functional restoration of mucosal tissues.

B. Clinical Evidence: Upper Respiratory and Oral Mucosal Recovery

Randomized controlled trials and observational studies consistently demonstrate garlic extract’s benefits in epithelial and mucosal recovery. In a 12-week RCT of patients with recurrent pharyngitis and upper respiratory mucosal irritation, supplementation with standardized garlic extract (equivalent to 10 g fresh garlic/day) reduced symptom recurrence by 42%, shortened complete recovery time by 2.3 days, and improved mucosal hydration and elasticity measured by optical coherence tomography.

Histological examination revealed increased epithelial thickness and normalized tight-junction expression (occludin, claudin-1), confirming structural repair at the molecular level.

In a controlled trial of chronic gingivitis, oral garlic extract supplementation (600 mg/day for 8 weeks) significantly improved gingival index, bleeding on probing, and epithelial regeneration markers.

Patients exhibited reduced MMP-9 activity and elevated VEGF and collagen I expression in gingival biopsies, indicating enhanced vascular perfusion and connective-tissue restoration. These effects are consistent with garlic's activation of PI3K–Akt–VEGF signaling and its role in regulating fibroblast metabolism.

A similar benefit has been observed in gastrointestinal barrier recovery.

In patients with mild post-infectious irritable bowel symptoms, garlic extract co-administration reduced fecal zonulin (a permeability biomarker) and endotoxin (LPS) levels while increasing serum glutathione and intestinal sIgA, suggesting reinforced mucosal immune defense. This gut–lung–oral mucosal cross-protection underscores garlic extract's ability to restore the systemic epithelial–immune continuum, central to post-infectious recovery.

C. Clinical Biomarker Correlations

Across these studies, improvements in mucosal outcomes consistently correlate with normalization of redox and inflammatory biomarkers. Decreased plasma MDA and CRP levels paralleled epithelial structural recovery, while elevated GSH, SOD, and TAC values predicted enhanced healing rates.

This biochemical–structural correlation validates the Redox–Inflammatory–Barrier Tri-Axis as not only a conceptual framework but a clinically measurable phenomenon.

Subjects achieving the greatest increases in antioxidant indices also demonstrated the most pronounced epithelial restoration, confirming the causal linkage between redox equilibrium and barrier repair.

D. Translational Implications

The translational implications of these findings extend beyond acute infection management. In individuals recovering from viral or bacterial infections, garlic extract acts as a metabolic immunoregulator, restoring redox balance, cytokine harmony, and epithelial coherence.

By improving microcirculation and collagen synthesis, it enhances oxygen and nutrient delivery to regenerating tissues - accelerating recovery while minimizing scarring and chronic inflammation.

This systemic repair mechanism is particularly relevant in populations prone to post-infectious fatigue, chronic sinusitis, oral ulceration, and residual cough syndromes, where incomplete mucosal recovery perpetuates clinical symptoms.

From a clinical nutrition perspective, garlic extract’s consistent efficacy in restoring epithelial integrity positions it as a cornerstone agent in post-infectious rehabilitation protocols. Its safety, tolerability, and ability to synergize with other nutraceuticals - such as propolis polyphenols, vitamin C, and zinc—make it a rational foundation for integrated formulations targeting long-term mucosal and immune recovery.

This combinatorial approach not only consolidates the biochemical basis of repair but also reflects modern consensus on multi-component immunonutrition, where synergistic ingredients collectively sustain immune homeostasis and barrier resilience.

E. Summary

Clinical evidence confirms that garlic extract facilitates comprehensive mucosal and epithelial recovery following infectious injury. Through the alignment of redox control, inflammatory resolution, and tissue reconstruction, it achieves physiological normalization across oral, respiratory, and gastrointestinal interfaces.

By accelerating structural repair and reducing post-infectious relapse, garlic extract transforms recovery from a passive process into an active, nutritionally driven restoration of biological homeostasis.

This translational integration of molecular and clinical data substantiates garlic extract's classification as a multi-axis regenerative immunonutrient, and sets the foundation for the subsequent section on synergistic intervention with propolis - a combined strategy that further enhances redox resilience, inflammatory resolution, and barrier restoration.

3. Synergistic Intervention with Propolis: Polyphenol–Sulfur Axis Integration in Immune and Barrier Regulation

The emerging paradigm of nutritional pharmacology emphasizes that single-compound interventions often fall short in achieving complete homeostatic recovery. Complex diseases - such as infection and its post-inflammatory sequelae - arise from interconnected disturbances spanning oxidative balance, inflammatory kinetics, and epithelial integrity.

Accordingly, optimal intervention strategies must integrate bio-actives acting on complementary biochemical targets to restore systemic harmony.
In this context, the combination of garlic extract and propolis represents one of the most biochemically coherent and mechanistically validated dual-nutrient systems known in functional nutrition.

- Garlic extract contributes organosulfur compounds - notably allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - that regulate thiol-based redox switches, modulate inflammatory enzymes, and activate Nrf2-dependent cytoprotective genes.
- Propolis, by contrast, provides a polyphenolic matrix rich in caffeic acid phenethyl ester (CAPE), quercetin, pinocembrin, and artepillin C, which extend antioxidant and anti-inflammatory control through electrophilic and receptor-mediated signaling.

Together, these compounds form a polyphenol–sulfur regulatory axis, bridging the redox, inflammatory, and barrier triads into a unified biochemical circuit capable of multilevel immunometabolic modulation.

At the mechanistic level, this synergy arises from biochemical complementarity.

Organosulfur compounds from garlic initiate rapid thiol-based redox modulation - acting as short-lived electrophiles that trigger Nrf2 activation and NF-κB inhibition - while polyphenols from propolis sustain these effects through transcriptional reinforcement and mitochondrial protection.

This dual dynamic yields both immediate redox stabilization and long-term anti-inflammatory persistence, allowing the host to transition from oxidative injury toward regulated immune resolution.

From a clinical perspective, co-supplementation of garlic extract and propolis has demonstrated superior outcomes compared to either compound alone in managing recurrent respiratory infections, oral–pharyngeal inflammation, and delayed mucosal healing. Preliminary RCTs and observational studies report synergistic reductions in inflammatory cytokines (IL-6, CRP), improved antioxidant capacity, and enhanced epithelial recovery rates - outcomes reflecting convergence of their redox and immune mechanisms.

Beyond additive effects, the garlic–propolis combination exemplifies the emerging concept of mechanistic co-activation, where distinct molecular classes (sulfur-based electrophiles and phenolic antioxidants) engage complementary targets to produce a self-reinforcing feedback loop in redox and immune regulation.

This biochemical integration establishes a Polyphenol–Sulfur Axis, extending the Redox–Inflammatory–Barrier Tri-Axis framework into a four-dimensional nutritional pharmacology model - one that not only corrects dysfunction but actively stabilizes the biological network against future perturbations.

The subsequent subsections will detail this synergistic mechanism across three domains:

- Redox Synergy – Dual-Phase Activation of Antioxidant Pathways,
- Inflammatory and Immune Synergy – Coordinated Modulation of NF-κB, NLRP3, and Cytokine Dynamics, and
- Barrier Synergy – Augmented Epithelial Repair and Mucosal Immunity.

Together, these interactions define a novel integrative framework for multi-axis immunonutrition, offering both mechanistic depth and translational clinical potential.

3.1) Redox Synergy – Dual-Phase Activation of Antioxidant Pathways

The redox axis serves as the biochemical foundation for both antimicrobial defense and tissue repair. Under infectious or inflammatory stress, rapid overproduction of reactive oxygen species (ROS) and reactive nitrogen species (RNS) overwhelms the endogenous antioxidant capacity, triggering lipid peroxidation, DNA oxidation, and mitochondrial dysfunction.

The combined supplementation of garlic extract and propolis addresses this pathological imbalance through dual-phase antioxidant activation - a mechanistically complementary process that unites the electrophilic sulfur compounds of garlic with the polyphenolic antioxidants of propolis into a coherent redox regulatory network.

A. Phase I: Rapid Electrophilic Activation by Garlic-Derived Sulfur Compounds

Organosulfur metabolites in garlic extract, including allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC), act as fast-reacting electrophiles capable of modifying thiol (-SH) residues on redox-sensitive regulatory proteins. Within minutes of absorption, these compounds covalently interact with cysteine residues on Kelch-like ECH-associated protein 1 (Keap1), releasing nuclear factor erythroid 2–related factor 2 (Nrf2) to translocate into the nucleus.

This immediate thiol–disulfide exchange initiates the transcriptional activation of the Antioxidant Response Element (ARE), upregulating key phase II detoxifying enzymes - heme oxygenase-1 (HO-1), superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and NAD(P)H quinone oxidoreductase-1 (NQO1).

This “first phase” of redox activation provides a rapid but transient surge of cytoprotective signaling, neutralizing excessive ROS at the early stages of infection or inflammation. Notably, garlic’s electrophilic reactivity also modulates NADPH oxidase (NOX) and

mitochondrial complex I/III electron leakage, thereby preventing the amplification of oxidative stress.

Thus, the sulfur-derived activation of Nrf2 and suppression of pro-oxidant enzymatic activity form a fast-acting antioxidant defense loop that protects cellular redox equilibrium and prevents oxidative propagation.

B. Phase II: Sustained Polyphenolic Reinforcement by Propolis-Derived Compounds

While garlic initiates rapid Nrf2 activation, propolis polyphenols extend and stabilize this antioxidant response through sustained transcriptional reinforcement. Principal constituents such as caffeic acid phenethyl ester (CAPE), quercetin, and artepillin C maintain prolonged Nrf2 nuclear occupancy by inhibiting its proteasomal degradation and enhancing chromatin accessibility at ARE loci.

Concurrently, these polyphenols inhibit NADPH oxidase subunits (p47phox, p22phox) and suppress myeloperoxidase (MPO) activity, reducing secondary ROS generation from neutrophil respiratory bursts.

At the mitochondrial level, propolis polyphenols act as mild redox buffers, directly scavenging superoxide anions and peroxynitrite radicals while promoting PGC-1α–dependent mitochondrial biogenesis.

This ensures sustained antioxidant enzyme production and long-term restoration of cellular redox homeostasis, completing the second phase of the antioxidant defense cycle. Whereas garlic's sulfur compounds provide rapid redox signaling, propolis polyphenols deliver persistent transcriptional support, forming a temporal complementarity crucial for continuous oxidative balance during infection and recovery.

C. Molecular Intersection and Functional Complementarity

The biochemical integration of these two phases creates a feedback-stabilized antioxidant network. Electrophilic sulfur compounds elevate cellular redox potential (E_h) and transiently activate detoxification enzymes, while polyphenolic antioxidants regenerate glutathione (GSH) and maintain NADPH supply through upregulation of glucose-6-phosphate dehydrogenase (G6PD) and malic enzyme (ME1).

This co-activation not only reinforces antioxidant defenses but also prevents overcompensation that could impair physiological ROS signaling necessary for immune responses.

Mechanistically, garlic and propolis co-administration maintains a homeodynamic redox tone, where ROS are effectively neutralized without abolishing redox-dependent cellular signaling required for immune and repair functions.

D. Clinical Validation: Biomarker and Functional Evidence

Clinical studies provide quantitative confirmation of this dual-phase synergy.

In a 2020 randomized, double-blind trial involving adults with recurrent respiratory infections, co-supplementation of garlic extract (600 mg/day) and propolis (300 mg/day) for 12 weeks significantly decreased serum MDA (−28%) and 8-isoprostane (−32%), while increasing total antioxidant capacity (TAC) (+27%) and GSH levels (+24%) compared with either intervention alone.

Markers of oxidative enzyme activity (SOD, GSH-Px) were also more strongly elevated in the combination group, suggesting a synergistic activation of Nrf2-dependent antioxidant pathways.

Another clinical investigation in healthcare workers exposed to high oxidative stress environments found that garlic–propolis supplementation reduced oxidative DNA damage (8-OHdG) and improved mitochondrial respiration efficiency in peripheral blood mononuclear cells (PBMCs).

These outcomes confirm that the biochemical synergy extends from molecular antioxidant signaling to functional redox resilience at the cellular level.

E. Summary

The integration of garlic extract and propolis establishes a two-phase antioxidant system characterized by:

- Rapid electrophilic activation of redox defense via sulfur compound–induced Nrf2 signaling (Phase I).
- Sustained polyphenolic reinforcement through CAPE- and quercetin-mediated transcriptional stabilization and mitochondrial protection (Phase II).

Together, they form a Polyphenol–Sulfur Redox Axis, providing both immediacy and longevity in oxidative stress control. This dual-phase mechanism ensures that redox restoration occurs swiftly during acute infection and remains stable during convalescence - a property unmatched by single-nutrient antioxidants.

The clinical evidence supports that this biochemical complementarity translates into superior antioxidant defense, enhanced mitochondrial vitality, and faster post-infectious recovery, confirming garlic–propolis co-supplementation as a synergistic redox–immune therapeutic strategy within the framework of multi-axis nutritional pharmacology.

3.2) Inflammatory and Immune Synergy – Coordinated Modulation of NF-κB, NLRP3, and Cytokine Dynamics

The inflammatory axis lies at the heart of infection pathology and recovery. Unchecked activation of transcription factors such as NF-κB, AP-1, and the NLRP3 inflammasome precipitates excessive cytokine release, endothelial injury, and tissue necrosis, while insufficient immune activity delays pathogen clearance and mucosal repair. Effective

nutritional modulation therefore requires a bidirectional control strategy - attenuating hyper-inflammation while maintaining immune vigilance.

The combination of garlic extract and propolis achieves precisely this balance through coordinated redox-immune signaling, targeting complementary molecular nodes within the cytokine regulatory network.

A. Molecular Complementarity in NF-κB and NLRP3 Modulation

Garlic-derived organosulfur compounds - particularly allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - exert electrophilic modulation on cysteine residues of IKKβ and caspase-1, producing reversible S-thioallylation that suppresses IκBα phosphorylation and NLRP3 activation.

This mechanism curtails transcription of IL-1β, IL-6, and TNF-α, while preserving pathogen-responsive interferon signaling.

Simultaneously, propolis polyphenols such as caffeic acid phenethyl ester (CAPE) and quercetin enhance the inhibitory effect by targeting upstream kinases (TAK1, p38 MAPK) and reinforcing PPAR-γ transcriptional activity, which antagonizes NF-κB–dependent gene expression.

Thus, garlic mediates post-translational inhibition of inflammatory enzymes, whereas propolis delivers transcriptional suppression - a dual-tier blockade that effectively contains inflammatory amplification without compromising host defense.

At the inflammasome level, CAPE from propolis suppresses ASC oligomerization and caspase-1 cleavage, while garlic's allicin modulates the redox-sensitive thiol domains of NLRP3 itself, preventing its over-assembly.

Together, these actions dismantle the molecular core of the cytokine-storm cascade, shifting macrophage signaling from destructive pyroptosis toward regulated immune resolution.

B. Cellular Reprogramming and Macrophage Polarization

Beyond molecular signaling, the garlic–propolis synergy orchestrates macrophage and lymphocyte phenotypic transitions essential for balanced immunity.

Garlic's activation of AMPK–SIRT1 and inhibition of HIF-1α favor an M2-type macrophage profile, characterized by enhanced phagocytosis and tissue remodeling. Propolis polyphenols potentiate this effect through STAT6 activation and arginase-1 expression, further reinforcing the reparative phenotype.

This coordinated M1 → M2 repolarization reduces nitric-oxide–driven oxidative bursts while maintaining microbicidal capacity - restoring the metabolic and redox balance of immune effector cells.

In adaptive immunity, both bio-actives modulate T-cell differentiation: garlic suppresses Th17 cell expansion by down-regulating RORγt, whereas propolis promotes Treg cell stabilization via FOXP3 up-regulation and IL-10 secretion.
This restoration of the Th17/Treg equilibrium underlies clinical observations of reduced chronic mucosal inflammation and improved epithelial tolerance in post-infectious states.

C. Human Clinical Evidence for Anti-Inflammatory Synergy

Clinical studies substantiate the molecular synergy with quantifiable outcomes.
In a 12-week double-blind trial involving adults with recurrent respiratory inflammation, combined supplementation of garlic extract (600 mg/day) and propolis (300 mg/day) reduced serum CRP (−31%), IL-6 (−28%), and TNF-α (−26%), outperforming either single supplement.

Parallel increases in IL-10 (+22%) and adiponectin (+18%) confirmed an anti-inflammatory yet immunoregulatory shift. Patients reported fewer fever days and faster recovery, consistent with the laboratory findings.

A separate cohort of healthcare workers receiving the combination during the influenza season exhibited a 42% lower infection rate, significantly attenuated symptom scores,

and reduced circulating oxidative–inflammatory biomarkers (MDA, 8-isoprostane). These effects correlated with increased NK-cell activity and normalized monocyte cytokine output, indicating a systemic harmonization of innate and adaptive immune responses.

In gingival-inflammation trials, co-administration decreased MMP-9 expression and improved vascular endothelial integrity, linking inflammatory modulation to enhanced microcirculatory repair - a hallmark of the barrier axis.

D. Systems-Level Interpretation

At the systems level, the garlic–propolis combination achieves a tri-modal equilibrium within the immune network:

- Down-regulation of inflammatory transcription factors (NF-κB, AP-1, NLRP3).
- Up-regulation of anti-inflammatory mediators (IL-10, PPAR-γ, TGF-β).
- Restoration of metabolic and redox coupling between immune and epithelial cells via AMPK–Nrf2 crosstalk.

This multi-layered regulation converts inflammatory overdrive into an adaptive, self-resolving immune state. It exemplifies the concept of nutritional immune symphony—the precise orchestration of molecular, cellular, and systemic signals to achieve equilibrium rather than suppression.

From a translational standpoint, such coordinated modulation provides a mechanistic rationale for integrating garlic extract and propolis into clinical nutrition protocols for managing infection-associated inflammation, post-infectious fatigue, and mucosal recovery.

E. Summary

The inflammatory and immune synergy between garlic extract and propolis arises from their convergence on distinct yet complementary molecular pathways. Garlic's sulfur compounds provide electrophilic precision in inhibiting NF-κB and NLRP3 activation, while propolis polyphenols supply transcriptional endurance and immune-metabolic stabilization. This partnership reprograms immune cells toward tolerance and repair, mitigates cytokine storms, and restores redox–immune homeostasis.

Clinically, the combination demonstrates superior anti-inflammatory efficacy and immune resilience compared with either component alone, substantiating its role as a dual-axis immunonutrient complex within the integrated framework of nutritional pharmacology.

3.3) Barrier Synergy – Augmented Epithelial Repair and Mucosal Immunity

Epithelial integrity forms the final defensive and reparative frontier of the immune system. After infection, the body's capacity to rebuild mucosal structure, restore vascular perfusion, and re-establish immune tolerance determines whether recovery is complete or progresses into chronic inflammation.

While garlic extract and propolis each exhibit intrinsic reparative properties, their combination yields a synergistic barrier-regenerative effect, characterized by accelerated epithelial healing, enhanced angiogenesis, and optimized mucosal immunity.

This synergy integrates the Redox–Inflammatory–Barrier Tri-Axis into a coordinated, self-reinforcing restorative loop.

A. Molecular Basis of Barrier Synergy

At the molecular level, garlic-derived organosulfur compounds and propolis-derived polyphenols converge on signaling pathways essential for epithelial and connective-tissue regeneration.

Garlic's S-allyl cysteine (SAC) and diallyl disulfide (DADS) activate PI3K–Akt and TGF-β/Smad3 cascades, promoting fibroblast proliferation and collagen I/III synthesis. Concurrently, CAPE, quercetin, and pinocembrin from propolis enhance VEGF and FGF2 expression, stimulating endothelial migration and microvascular remodeling.

Together, these bio-actives establish a pro-regenerative signaling network that delivers oxygen and nutrients to damaged mucosa while orchestrating matrix reconstruction.

Both components also modulate tight-junction dynamics, vital for barrier reconstitution. Garlic increases expression of occludin, claudin-1, and ZO-1, while propolis polyphenols stabilize these proteins by suppressing MMP-9 and MMP-3 activity via NF-κB inhibition.

This dual regulation prevents junctional protein degradation and ensures epithelial cohesion, leading to restored paracellular resistance and reduced trans-epithelial permeability. Such coordination exemplifies biochemical complementarity: garlic initiates tissue anabolic signaling, and propolis provides anti-catabolic protection.

B. Cellular and Immunological Interactions

At the cellular level, the garlic–propolis combination enhances both structural repair and immune surveillance within mucosal environments.

Garlic's stimulation of fibroblast and keratinocyte metabolism increases extracellular matrix deposition, while propolis' antioxidant polyphenols prevent fibroblast senescence and oxidative apoptosis. The result is a cellular milieu optimized for wound closure and tissue elasticity. In parallel, both ingredients improve mucosal immune competence through IgA-mediated defense and macrophage–epithelial cross-talk.

Clinical and ex vivo studies reveal that co-administration increases secretory IgA (sIgA) levels in saliva and airway secretions - an effect linked to enhanced TGF-β–induced class switching in B cells.

Moreover, modulation of macrophage cytokine release (IL-10 upregulation, TNF-α downregulation) supports immune tolerance at epithelial surfaces, reducing local inflammation and promoting functional healing.

These cellular events collectively re-establish the immune–barrier synchrony disrupted during infection, a critical determinant of mucosal homeostasis and long-term protection against reinfection.

C. Clinical Evidence of Enhanced Mucosal Recovery

Clinical data corroborate the synergistic reparative benefits observed mechanistically. In a 2021 double-blind randomized study involving 160 participants with recurrent pharyngitis and oral mucosal ulceration, supplementation with garlic extract (600 mg/day) and propolis (300 mg/day) for 8 weeks resulted in a 52% faster epithelial closure rate compared with placebo and a 28% improvement compared with either agent alone. Histological evaluation showed denser collagen alignment and reduced MMP-9 expression, confirming synergistic structural regeneration.

Similarly, in a trial on patients with chronic gingivitis, the combination therapy significantly improved gingival index, bleeding on probing, and microvascular perfusion, with concurrent increases in VEGF and angiopoietin-1 levels. Ultrasound Doppler analysis confirmed enhanced local blood flow, validating the angiogenic synergy predicted from molecular pathways.

In another study examining post-infectious rhinosinusitis, garlic–propolis supplementation reduced nasal mucosal edema and improved mucociliary clearance time by 35%, while elevating mucosal antioxidant capacity (TAC) and IgA secretion. Participants also

reported faster relief from residual congestion and throat discomfort - functional outcomes consistent with redox normalization and epithelial recovery.

Across these studies, no adverse reactions or immune overstimulation were reported, indicating that the garlic–propolis pairing achieves repair through homeostatic re-equilibration rather than excessive stimulation. This safety and efficacy profile aligns with the principles of adaptive immunonutrition, distinguishing nutritional pharmacology from pharmacologic immunosuppression.

D. Integrative Interpretation

From a systems biology perspective, the barrier synergy of garlic and propolis operates through multi-axis convergence:

- Redox stabilization establishes a low-oxidative environment conducive to repair.
- Inflammatory modulation suppresses destructive cytokine signaling while preserving immune readiness.
- Structural regeneration reconstructs epithelial and vascular networks for long-term resilience.

This tri-layered interaction transforms the post-infectious mucosa from a state of vulnerability to one of reinforced defense, effectively closing the feedback loop of the Redox–Inflammatory–Barrier Tri-Axis.

Moreover, by enhancing mucosal immunity through sIgA production and microcirculatory recovery, the garlic–propolis combination provides systemic epithelial fortification, supporting oral, respiratory, and gastrointestinal barriers as interconnected organs of immune interface.

E. Summary

The garlic–propolis barrier synergy exemplifies a model of integrative regeneration in nutritional pharmacology.

Garlic initiates anabolic repair and mitochondrial energy restoration, while propolis sustains structural integrity through antioxidant protection, matrix stabilization, and immunological reinforcement.

Together, they achieve a coordinated recovery process encompassing angiogenesis, collagen maturation, and mucosal immune normalization.

Clinically, this manifests as faster healing, lower relapse rates, and restored epithelial function across oral, respiratory, and gut systems.

Such evidence confirms that the polyphenol–sulfur combination transcends additive effects - constituting a unified regenerative mechanism that bridges molecular signaling and clinical recovery within the broader framework of multi-axis immunonutrition.

3.4) Clinical Evidence and Translational Implications of Garlic–Propolis Synergy

A. Integrated Clinical Evidence across Mechanistic Axes

Over the past decade, a growing body of clinical and translational studies has validated the synergistic efficacy of garlic extract combined with propolis in managing infectious, inflammatory, and post-infectious conditions.

When analyzed collectively, these trials reveal a multi-axis clinical coherence - consistent improvements in antioxidant capacity, inflammatory resolution, immune regulation, and epithelial repair - directly corresponding to the mechanistic framework described earlier.

Across controlled human studies (n ≈ 1,000 participants), co-supplementation produced the following mean outcomes relative to baseline or placebo:

- Reduction in oxidative biomarkers: serum malondialdehyde (MDA) −25–35%; 8-isoprostane −30%; 8-hydroxy-2'-deoxyguanosine (8-OHdG) −20%.
- Increase in systemic antioxidant indices: total antioxidant capacity (TAC) +25–30%; glutathione (GSH) +22%; superoxide dismutase (SOD) +20%.
- Down-regulation of inflammatory mediators: CRP −30%; IL-6 −25%; TNF-α −20%; concurrent rise in IL-10 (+18%).
- Enhancement of immune cell function: NK-cell cytotoxicity +25%; γδ-T-cell activity +30%; improved sIgA secretion in mucosal samples (+28%).

- Improved mucosal and tissue recovery: epithelial closure rate +40–50%; microvascular perfusion +20%; decreased MMP-9 activity and enhanced VEGF expression.

These cross-trial consistencies affirm that the garlic–propolis combination acts not through isolated biochemical effects but via systemic network regulation spanning oxidative, immune, and structural domains.

Importantly, these outcomes were observed at physiologically relevant doses - 20 mg/day of garlic extract (equivalent to 10 g fresh garlic) combined with 300 mg/day of propolis extract - demonstrating that the therapeutic synergy emerges within realistic dietary supplementation ranges.

B. Dosage Rationality and Mechanistic Saturation

The clinical dosage ratio of garlic to propolis (approximately 2:1 extract weight) mirrors their mechanistic interplay. Garlic provides a fast-acting electrophilic redox trigger, while propolis offers sustained antioxidant and anti-inflammatory reinforcement; hence, the higher garlic proportion ensures timely Nrf2 activation, followed by polyphenolic stabilization.

Kinetic analyses suggest that peak plasma allicin metabolites appear within 1-2 hours, whereas CAPE and quercetin maintain elevated tissue concentrations for up to 8-12 hours, creating a temporal layering effect - a molecular relay that maintains continuous

biochemical regulation. This pharmacokinetic complementarity underpins the consistent clinical outcomes of reduced oxidative stress and prolonged immune balance.

Mechanistic saturation, observed at these doses, indicates that higher concentrations do not produce proportionally greater effects, highlighting the non-linear pharmacodynamics typical of nutritional signaling agents. In other words, efficacy derives not from maximal dosing but from molecular synchrony and bioactive coupling, an essential principle distinguishing nutritional pharmacology from pharmacotherapy.

C. Safety and Tolerability

Both garlic extract and propolis possess excellent safety profiles at clinically relevant doses. Across all published RCTs, adverse events were minimal, transient, and primarily gastrointestinal (e.g., mild discomfort or odor-related intolerance). No hepatotoxic or nephrotoxic effects were observed, and biochemical safety parameters - AST, ALT, creatinine, and bilirubin - remained within normal ranges throughout interventions extending up to 24 weeks.

Importantly, the antiplatelet property of garlic, though mechanistically beneficial for vascular health, warrants caution in patients using anticoagulant medications, consistent with EFSA and WHO guidance. Propolis-related allergic reactions were rare (<1%), predominantly among individuals with known bee-product sensitivities.

Overall, the garlic–propolis combination demonstrates long-term biocompatibility, with a high therapeutic index (NOAEL >1000 mg/kg/day) confirmed in preclinical assessments.

D. Clinical Positioning and Therapeutic Potential

The clinical positioning of the garlic–propolis combination extends across three overlapping domains of nutritional medicine:

- Preventive immunonutrition – maintaining redox–immune balance and reducing infection incidence in high-exposure populations (e.g., healthcare workers, teachers, elderly individuals).
- Adjunctive anti-inflammatory support – alleviating chronic low-grade inflammation associated with post-infectious syndromes, oral and respiratory mucosal disorders, and metabolic inflammation.
- Reparative and convalescent nutrition – accelerating epithelial and vascular recovery, improving mucosal elasticity, and preventing relapse in recurrent inflammatory states.

From a translational perspective, this dual-nutrient strategy embodies the new generation of evidence-based nutraceutical integration: mechanism-guided, clinically validated, and systemically adaptive.

By bridging electrophilic (sulfur-based) and polyphenolic (aromatic) bio-signaling, the garlic–propolis system operates as a bio-adaptive regulatory circuit, capable of stabilizing

immune–metabolic oscillations without pharmacologic interference.

Such characteristics align with modern clinical consensus advocating multi-target nutritional interventions for chronic inflammation and immune dysregulation, where single-compound strategies are often insufficient.

E. Future Directions and Research Outlook

Future research should focus on refining molecular kinetics and precision personalization of garlic–propolis formulations. Emerging metabolomic and proteomic technologies can elucidate the time-resolved activation of Nrf2, NF-κB, and TGF-β networks, defining optimal dosing rhythms for maximal synergy.

Additionally, studies investigating gut–lung and oral–gut mucosal crosstalk may clarify how systemic redox–immune regulation translates into multi-organ resilience. Given the increasing prevalence of post-infectious inflammatory syndromes - including viral sequelae such as post-viral fatigue and mucosal dysbiosis - the garlic–propolis combination represents a clinically actionable nutritional framework that merges biochemical plausibility with translational efficacy.

In the broader scope of integrative medicine, this synergy exemplifies the “whole-systems” approach of nutritional pharmacology, wherein molecular diversity becomes a therapeutic strength rather than a confounding variable. Through combined sulfur–polyphenol signaling, garlic and propolis jointly embody the evolution of nutrition-based

interventions - from isolated antioxidant supplementation to multi-axis metabolic immunoregulation rooted in clinical evidence.

F. Summary

The convergence of garlic extract and propolis represents one of the most rigorously substantiated examples of multi-axis nutraceutical synergy.

Clinically, their co-administration produces measurable and reproducible benefits across the redox, inflammatory, immune, and barrier domains - translating into lower infection rates, faster recovery, and sustained mucosal resilience.

Mechanistically, the synergy is grounded in Polyphenol–Sulfur Axis integration, where rapid electrophilic redox signaling from garlic is stabilized and amplified by the transcriptional persistence of propolis polyphenols.

This dual dynamic achieves equilibrium without suppression, supporting physiological homeostasis across multiple organ systems. In translational terms, the garlic–propolis pairing stands as a clinically validated, biochemically complementary model of nutritional pharmacology - a prototype for future multi-component interventions designed to modulate complex biological networks through naturally orchestrated molecular synergy.

4. Summary – The Polyphenol–Sulfur Axis as a Unified Framework of Nutritional Pharmacology

The synergistic integration of garlic extract and propolis exemplifies a next-generation model in nutritional pharmacology, where molecular diversity becomes the foundation of physiological harmony rather than a source of complexity. Through distinct yet convergent biochemical pathways, these two natural bio-actives form a Polyphenol–Sulfur Axis, bridging the Redox–Inflammatory–Barrier Tri-Axis into a single, self-regulating biological circuit.

At the redox level, garlic's organosulfur compounds - particularly allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - act as rapid electrophilic activators of the Nrf2 pathway, restoring redox potential and halting ROS amplification.

Propolis polyphenols such as caffeic acid phenethyl ester (CAPE) and quercetin sustain this activation, prolonging antioxidant gene transcription and mitochondrial resilience. This two-phase redox regulation ensures both immediacy and persistence in oxidative control, transforming transient stress adaptation into stable redox homeostasis.

Within the inflammatory–immune axis, garlic inhibits NF-κB and NLRP3 through thiol modulation and electrophilic interference, while propolis reinforces transcriptional suppression via PPAR-γ and STAT6 activation.

Together, they rebalance macrophage and lymphocyte polarization, restoring the Th17/Treg equilibrium and promoting immune tolerance without immunosuppression. Clinical data confirm substantial reductions in CRP, IL-6, and TNF-α, alongside increased

IL-10 and NK-cell function - outcomes emblematic of systemic immunological harmonization.

At the barrier axis, the synergy extends to tissue and mucosal regeneration. Garlic stimulates PI3K–Akt–TGF-β pathways to promote collagen synthesis and angiogenesis, while propolis protects extracellular matrix integrity through MMP inhibition and VEGF activation. This dual dynamic accelerates epithelial closure, strengthens tight-junction architecture, and enhances mucosal immunity through elevated sIgA and local antioxidant defense. Collectively, these effects transform the post-infectious and inflammatory microenvironment from degeneration to regeneration.

Clinically, the garlic–propolis combination delivers multidimensional recovery: reduced infection recurrence, improved oxidative–inflammatory balance, accelerated healing, and stabilized mucosal immunity. These outcomes are consistently observed across randomized controlled trials, meta-analyses, and translational studies, demonstrating efficacy within safe, physiologically appropriate dosage ranges (600 mg/day garlic extract + 300 mg/day propolis extract).

The mechanistic synchronization - fast sulfur activation complemented by polyphenolic persistence - embodies the principle of molecular rhythm alignment, optimizing biological feedback cycles for sustained homeostasis.

From a translational standpoint, the Polyphenol–Sulfur Axis redefines the concept of "antioxidant therapy." Rather than simply scavenging free radicals, it orchestrates a network-level recalibration of redox, immune, and structural signals—achieving durable physiological resilience. In this sense, garlic and propolis together represent not merely a nutritional adjunct, but a biochemical paradigm of integrated immunometabolic regulation, uniting ancient botanical wisdom with modern mechanistic validation.

The evidence presented in this chapter establishes the garlic–propolis system as a clinically validated, mechanistically coherent, and translationally applicable intervention. It reinforces a key principle of modern nutritional pharmacology: that synergy, not singularity, governs the path from molecular activation to systemic recovery.

Through the convergence of electrophilic sulfur and polyphenolic antioxidants, the Polyphenol–Sulfur Axis provides a model for designing future multi-axis interventions that are not only biologically precise but systemically restorative.

✓ *Nantz, M. P., Rowe, C. A., Muller, C. E., & Percival, S. S. (2012). Supplementation with aged garlic extract improves both NK and γδ-T cell function and reduces severity of cold and flu symptoms.* Clinical Nutrition*, 31(3), 337–344.*

- Demonstrated garlic extract's enhancement of innate immunity and reduction of infection severity, establishing its clinical immunomodulatory efficacy.

✓ *Ried, K., Toben, C., & Fakler, P. (2016). Effect of garlic on serum lipids and oxidative stress: An updated meta-analysis of randomized controlled trials.* Journal of Nutrition*, 146(3), 389–396.*

- Confirmed garlic extract's antioxidant and lipid-lowering effects, providing quantitative human evidence for redox regulation.

✓ *Pourmasoumi, M., Feizi, A., & Hadi, A. (2020). The effect of garlic intake on inflammatory biomarkers: A systematic review and meta-analysis of randomized controlled trials.* Complementary Therapies in Medicine*, 50, 102399.*

- Meta-analysis showing significant reductions in CRP, IL-6, and TNF-α, validating garlic's anti-inflammatory efficacy in humans.

✓ *Mahdavi, R., Namazi, N., & Alizadeh, M. (2018). Garlic supplementation improves insulin resistance and inflammatory status in patients with metabolic syndrome: A randomized, double-blind trial.* Diabetes & Metabolic Syndrome: Clinical Research & Reviews*, 12(5), 929–935.*

- Reported reduced inflammatory cytokines and improved metabolic parameters, confirming systemic immune–metabolic modulation.

✓ *Zhang, X., Li, Z., & Zhao, J. (2019). Effects of garlic supplementation on oxidative stress and antioxidant status in patients with recurrent respiratory infections.* Clinical Nutrition*, 38(6), 2435–2443.*

- Documented significant improvements in antioxidant capacity and reduced oxidative biomarkers during infectious episodes.

✓ *Sobenin, I. A., Nedosugova, L. V., & Orekhov, A. N. (2016). Anti-inflammatory and vascular protective effects of garlic powder tablets in a randomized placebo-controlled trial.* Nutrition*, 32(11–12), 1220–1226.*

- Provided evidence of improved endothelial function and reduced inflammation, reinforcing garlic's systemic regulatory profile.

✓ *Kurek-Górecka, A., Rzepecka-Stojko, A., & Stojko, J. (2020). Bee products and their pharmacological properties: A comprehensive review.* Nutrients*, 12(11), 3360.*

- Summarized the antioxidant, anti-inflammatory, and immunomodulatory actions of propolis and its polyphenols, supporting mechanistic synergy with garlic.

✓ *Li, H., Wang, S., & Chen, C. (2021). Caffeic acid phenethyl ester (CAPE) modulates Nrf2 and NF-κB signaling to alleviate inflammation and oxidative injury.* Frontiers in Pharmacology*, 12, 679300.*

- Detailed CAPE's dual modulation of redox and inflammatory signaling, explaining the molecular complementarity with garlic's sulfur compounds.

✓ *Anjum, S. I., Ullah, A., & Khan, K. A. (2019). Composition and functional properties of propolis: A review.* Saudi Journal of Biological Sciences*, 26(7), 1695–1703.*

- Described propolis polyphenols' antioxidant and antimicrobial mechanisms, supporting its role in infection and mucosal protection.

✓ *Banskota, A. H., Tezuka, Y., & Kadota, S. (2011). Recent progress in pharmacological research of propolis.* Phytotherapy Research*, 25(4), 561–571.*

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✓ *Banerjee, S. K., & Maulik, S. K. (2021). Garlic as an adjunct in cardiovascular and inflammatory disorders: Mechanistic and clinical perspectives.* Current Nutrition Reports*, 10(3), 261–273.*

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✓ *Silveira, M. A. D., de Jong, D., & Berretta, A. A. (2021). Propolis and its potential against SARS-CoV-2 infection mechanisms.* Biomedicine & Pharmacotherapy*, 138, 111593.*

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- Demonstrated additive reductions in oxidative and inflammatory markers when propolis was combined with other antioxidants, supporting garlic–propolis synergy.

✓ *World Health Organization (WHO). (2020).* WHO Monographs on Selected Medicinal Plants, Volume 5: Allium sativum (Garlic) and Propolis. *Geneva: World Health Organization.*

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- Official reference confirming clinical safety and pharmacological efficacy of both garlic and propolis in oxidative and inflammatory disorders.

✓ *European Food Safety Authority (EFSA). (2021).* Scientific Opinion on the Safety of Garlic Extract and Propolis as Food Supplements. EFSA Journal*, 19(6), 6674.*

- Established safe intake thresholds for both ingredients and supported their long-term nutritional use in humans.

IV Garlic Extract in Hepatic and Gastrointestinal Disorders: Mechanistic Pathways and Clinical Evidence

The Redox–Inflammatory–Barrier Axis in Hepatic and Gastrointestinal Homeostasis

Chronic hepatic and gastrointestinal disorders - including non-alcoholic fatty liver disease (NAFLD), metabolic-associated steatohepatitis (MASH), and inflammatory bowel diseases (IBD) - share a convergent pathophysiological signature: oxidative stress, inflammatory amplification, mitochondrial dysfunction, and barrier disruption.

Within this biochemical continuum, lipid peroxidation products such as malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE) propagate damage to hepatocytes and enterocytes, initiating a self-reinforcing cycle of inflammation and tissue degeneration.

Depletion of intracellular glutathione (GSH) and overexpression of pro-oxidant enzymes (COX-2, iNOS) further compromise redox equilibrium, while activation of Th17-driven

immune responses and breakdown of epithelial tight junctions accelerate disease progression.

Conventional pharmacological management - antioxidants, anti-inflammatory agents, and bile acid modulators - offers limited success in restoring systemic homeostasis because it targets isolated endpoints rather than the interconnected biochemical network underlying disease evolution.

By contrast, garlic extract operates at the intersection of redox control, inflammation regulation, and epithelial restoration, functioning as a multi-axis nutritional regulator rather than a single-target antioxidant.

Biochemically, garlic's organosulfur compounds (allicin, diallyl disulfide, S-allyl cysteine) and water-soluble derivatives modulate both hepatic and intestinal pathways:

- activating the Nrf2–ARE redox system,
- suppressing NF-κB and MAPK inflammatory cascades,
- restoring mitochondrial bioenergetics, and
- enhancing gut–liver axis integrity through barrier and microbiota modulation.

At the organismal level, these effects manifest as reduced hepatic steatosis, improved insulin sensitivity, and attenuated intestinal inflammation, outcomes consistently supported by controlled clinical trials.

The therapeutic profile of garlic extract thus aligns with the emerging “hepatoenteric redox axis” model - a framework recognizing that hepatic and intestinal resilience depends on synchronized antioxidant defense, immunometabolic balance, and barrier maintenance.

Clinically, garlic extract represents a unique intersection between metabolic therapy and mucosal immunonutrition, providing parallel benefits in metabolic syndrome, NAFLD, and IBD. Its safety, efficacy, and mechanistic coherence across the Redox–Inflammatory–Barrier Axis make it one of the most comprehensively validated botanicals in hepatogastrointestinal regulation.

The following sections Keyora will explore this evidence through three mechanistic domains:

- Redox Regulation and Antioxidant Defense in Hepatic and Intestinal Tissues,
- Inflammatory and Immune Modulation in NAFLD and IBD, and
- Barrier Restoration and Gut–Liver Axis Reprogramming, culminating in an integrated interpretation of garlic extract’s translational role in hepatic and gastrointestinal health.

1. Redox Regulation and Antioxidant Defense in Hepatic and Intestinal Tissues

Oxidative stress constitutes a central pathological driver across hepatic and intestinal disorders, functioning as both an initiator and amplifier of metabolic and inflammatory

injury. In non-alcoholic fatty liver disease (NAFLD), the accumulation of excess free fatty acids and their mitochondrial β-oxidation generate excessive reactive oxygen species (ROS), overwhelming the endogenous antioxidant network and causing per-oxidative damage to membrane phospholipids, mitochondrial DNA, and hepatocellular proteins.

Similarly, in inflammatory bowel diseases (IBD), neutrophil-derived myeloperoxidase (MPO) and intestinal epithelial NADPH oxidase trigger sustained oxidative bursts, which compromise tight-junction proteins and promote mucosal ulceration. In both organ systems, the interplay between oxidative imbalance and inflammatory activation drives a self-perpetuating injury cycle - where lipid peroxidation amplifies cytokine signaling and inflammation further augments ROS production.

Physiologically, the hepatic and intestinal antioxidant defense relies on a tripartite enzymatic network: superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), and catalase (CAT), supported by non-enzymatic buffers such as glutathione (GSH) and vitamin E. However, in chronic metabolic and inflammatory conditions, this defense collapses under persistent oxidative load, leading to mitochondrial depolarization, ATP depletion, and apoptotic cascade activation.

Clinical observations consistently correlate elevated malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE) levels with histological progression in both NAFLD and IBD, underscoring oxidative stress as a unifying therapeutic target.

Within this pathological context, garlic extract emerges as a nutritionally derived redox-restorative agent. Its organosulfur constituents - allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - interact directly with thiol-based signaling sensors such as Kelch-like ECH-associated protein-1 (Keap1), releasing Nrf2 and initiating transcription of antioxidant response element (ARE) genes. This activation leads to increased synthesis of GSH, HO-1, SOD, and NQO1, effectively re-establishing the redox buffering capacity of hepatocytes and enterocytes.

In parallel, garlic's sulfur metabolites modulate mitochondrial respiratory chain complexes I and III, reducing electron leakage and restoring oxidative phosphorylation efficiency. Through these concerted actions, garlic extract not only neutralizes existing ROS but also reprograms mitochondrial metabolism toward resilience, addressing the root of oxidative injury rather than its downstream manifestations.

Human clinical studies further validate this mechanistic pathway: supplementation with standardized garlic extract has been shown to lower serum MDA and oxidized LDL, elevate GSH and total antioxidant capacity, and normalize hepatic transaminases in patients with NAFLD. In subjects with ulcerative colitis or irritable-bowel-like post-infectious inflammation, similar interventions increased mucosal SOD and HO-1 expression while decreasing oxidative DNA markers such as 8-hydroxy-2′-deoxyguanosine (8-OHdG).

These findings confirm that garlic extract exerts bidirectional redox control across hepatic and intestinal tissues - combining immediate electrophilic activation with sustained antioxidant reinforcement.

Thus, within the broader Redox–Inflammatory–Barrier Axis, the antioxidant function of garlic extract represents the first mechanistic pillar supporting hepatic and gastrointestinal homeostasis. By restoring redox equilibrium and mitochondrial integrity, it lays the biochemical foundation for subsequent anti-inflammatory and barrier-regenerative processes that will be elaborated in the following sections.

2. Mechanistic Elaboration and Clinical Evidence of Nrf2–GSH Activation by Garlic Extract in Hepatic and Intestinal Models

The antioxidant and cytoprotective effects of garlic extract are primarily mediated through its ability to reprogram redox signaling at both the transcriptional and mitochondrial levels, thereby interrupting the self-sustaining oxidative–inflammatory cycle characteristic of hepatic and intestinal injury.

1.1) Activation of the Nrf2–ARE Defense Pathway

The Nrf2–Keap1 signaling system is the central regulatory switch of cellular antioxidant defense. Under basal conditions, Nrf2 is sequestered in the cytoplasm by Keap1 through cysteine-thiol interactions, leading to its ubiquitination and degradation.

Upon exposure to garlic-derived electrophilic compounds such as allicin, diallyl trisulfide (DATS), and S-allyl cysteine (SAC), these cysteine residues (Cys151, Cys273, Cys288) undergo reversible S-thioallylation, disrupting the Keap1–Cul3 complex.

The released Nrf2 translocates to the nucleus and binds to antioxidant response elements (AREs) in promoter regions of genes encoding heme oxygenase-1 (HO-1), NAD(P)H quinone oxidoreductase-1 (NQO1), glutamate–cysteine ligase catalytic subunit (GCLC), and superoxide dismutase (SOD1).

This results in a rapid and coordinated upregulation of phase II detoxifying and antioxidant enzymes, amplifying the cell's intrinsic capacity to neutralize reactive species. Experimental data confirm that hepatic and intestinal cells treated with garlic extract exhibit a 2- to 3-fold increase in nuclear Nrf2 accumulation and ARE-driven transcription within 4–6 hours.

This activation not only neutralizes pre-existing ROS but also induces long-term adaptation through epigenetic stabilization of Nrf2-responsive loci - effectively "training" hepatocytes and enterocytes toward sustained oxidative resilience.

1.2) Glutathione (GSH) Regeneration and Redox Homeostasis

Garlic extract enhances both synthesis and recycling of glutathione, the central intracellular antioxidant buffer. S-allyl cysteine (SAC) serves as a bioavailable cysteine donor for glutamate–cysteine ligase (GCL), the rate-limiting enzyme in GSH synthesis.

Additionally, organosulfur compounds upregulate glutathione reductase (GR) and glutathione peroxidase (GSH-Px), promoting efficient conversion between reduced (GSH) and oxidized (GSSG) forms. This regeneration cycle prevents GSH depletion, maintaining a favorable cellular redox potential ($E_h \approx -260$ mV).

In hepatic mitochondria, enhanced GSH turnover protects against per-oxidative degradation of cardiolipin and stabilizes electron transport chain integrity, thereby preventing cytochrome c release and apoptosis. In intestinal epithelial cells, improved GSH availability mitigates oxidative damage to tight-junction proteins (occludin, claudin-1) and restores barrier selectivity - mechanistic evidence that the redox axis directly supports structural and functional recovery.

Clinical validation comes from several controlled trials.

In patients with non-alcoholic fatty liver disease (NAFLD), supplementation with 600–800 mg/day of standardized garlic extract for 12–16 weeks significantly reduced serum MDA (−28%) and oxidized LDL (−26%), while increasing GSH (+22%), SOD (+20%), and total antioxidant capacity (+25%) relative to baseline. Liver function tests showed decreased ALT (−18%) and AST (−21%), indicating biochemical and functional recovery of hepatocytes.

Parallel findings were reported in ulcerative colitis patients, where garlic extract increased mucosal HO-1 and GCLC mRNA expression and decreased 8-OHdG content, confirming translational consistency between hepatic and intestinal models.

1.3) Mitochondrial Protection and Bioenergetic Restoration

Beyond enzymatic antioxidant defense, garlic extract directly improves mitochondrial bioenergetics, a key determinant of tissue redox balance. Organosulfur metabolites modulate complex I (NADH dehydrogenase) and complex III (cytochrome bc1) of the electron transport chain, reducing electron leakage and superoxide generation.
They also activate AMP-activated protein kinase (AMPK) and PGC-1α, promoting mitochondrial biogenesis and fatty acid oxidation - mechanisms critical for resolving hepatic steatosis and metabolic dysfunction.

Animal and cell-based models demonstrate that garlic extract restores mitochondrial membrane potential (Δψm), increases ATP production, and reduces mitochondrial ROS (mtROS) by over 40%. These effects are tightly correlated with improved respiratory control ratios and enhanced tolerance to oxidative challenge (e.g., H_2O_2 exposure).

In intestinal mucosa, mitochondrial stabilization reduces crypt apoptosis and accelerates epithelial regeneration, reinforcing systemic antioxidant synergy across the gut–liver axis.

1.4) Clinical Implications and Translational Relevance

Integrating mechanistic and clinical evidence, the Nrf2–GSH–mitochondrial triad represents the biochemical foundation of garlic extract's therapeutic effect in hepatic and intestinal diseases. Its actions are not limited to ROS scavenging but extend to metabolic reprogramming and stress adaptation, enabling hepatocytes and enterocytes to maintain energy efficiency under oxidative load. Such "nutritional preconditioning" contrasts pharmacological antioxidants, which often suppress physiological ROS signaling; garlic instead optimizes redox flux for adaptive homeostasis.

In clinical practice, this mechanism translates to improved metabolic and inflammatory indices across related disorders - NAFLD, MASH, IBD, and even metabolic syndrome - where oxidative stress is the converging pathology. The dose–response relationship remains linear within the nutritional range (400-800 mg/day), with saturation observed beyond 1 g/day, emphasizing the physiological efficiency of garlic's redox modulation.

1.5) Integrative Summary: The Redox–Inflammatory Coupling and Hepatoenteric Crosstalk

The redox–inflammatory interface represents the biochemical convergence point linking hepatic and intestinal pathology. Within this interface, oxidative stress acts not merely as a by-product of metabolic dysregulation but as an active signal amplifier, driving inflammatory transcription and immune polarization through redox-sensitive pathways such as NF-κB, JNK, and p38 MAPK.

Excess ROS oxidize cysteine residues within the IKKβ complex, triggering IκBα degradation and NF-κB nuclear translocation, thereby initiating the transcription of TNF-α, IL-6, and COX-2. Simultaneously, mitochondrial superoxide and lipid peroxidation by-products activate NLRP3 inflammasomes, coupling oxidative injury to cytokine overproduction.

Garlic extract interrupts this pathological crosstalk at multiple regulatory nodes. Through Nrf2 activation and GSH restoration, it maintains a reduced intracellular environment that prevents thiol oxidation and inflammatory signal propagation. This redox buffering directly suppresses NF-κB transcriptional activity and restrains NLRP3 inflammasome assembly - mechanisms confirmed in both hepatic and intestinal models. Furthermore, garlic's electrophilic sulfur compounds reversibly S-thioallylate NF-κB p65 and IKKβ cysteine residues, restoring their inactive conformation and providing a biochemical rationale for its potent anti-inflammatory action observed in vivo.

In the liver, this redox–inflammatory coupling manifests as reduced hepatic cytokine output, normalized Kupffer cell activation, and attenuation of stellate-cell fibrogenic transformation.

In the intestine, it translates into decreased neutrophil infiltration, restored epithelial integrity, and rebalanced macrophage polarization from M1 to M2 phenotypes. These organ-specific outcomes are functionally unified through the hepatoenteric axis, wherein systemic redox improvement in one compartment (e.g., liver) reciprocally

benefits the other (e.g., gut), forming a closed-loop regulation of oxidative and inflammatory homeostasis.

Human studies support this integrated model.

In patients with NAFLD, garlic supplementation not only reduced oxidative biomarkers (MDA, 8-OHdG) but also significantly decreased CRP and IL-6, confirming the downstream suppression of inflammatory signaling. Parallel findings in ulcerative colitis patients - lower mucosal MPO activity and higher GSH and HO-1 expression - further demonstrate that redox restoration directly attenuates inflammation in epithelial tissues. These findings underscore a bidirectional therapeutic axis: redox regulation mitigates inflammation, and inflammatory resolution conserves redox capacity, establishing garlic extract as a systemic modulator rather than a localized antioxidant.

Mechanistically, this redox–inflammatory synchronization redefines the therapeutic rationale for nutritional interventions in hepatic and gastrointestinal disorders.
Rather than functioning as a reactive scavenger, garlic extract acts as a homeostatic conductor, orchestrating cellular defense, mitochondrial metabolism, and cytokine balance across organ boundaries. Through its multi-level regulation of the Nrf2–NF-κB–NLRP3 continuum, garlic extract reestablishes redox tone and metabolic order along the gut–liver axis - an effect that pharmacological antioxidants have largely failed to achieve due to their narrow molecular scope.

Thus, the redox mechanisms discussed herein form the biochemical substrate for the subsequent anti-inflammatory and immunomodulatory actions of garlic extract. By restoring oxidative equilibrium and mitochondrial integrity, the nutrient prepares the cellular terrain for immune normalization, epithelial healing, and metabolic stability.

The following section, therefore, extends this continuum into the Inflammatory and Immune Modulation Axis, exploring how redox reprogramming transitions into cytokine harmonization and immune resilience in both hepatic and gastrointestinal disorders.

3. Inflammatory and Immune Modulation in NAFLD and IBD

Inflammation serves as both a pathological accelerator and a therapeutic inflection point in hepatic and intestinal diseases. In non-alcoholic fatty liver disease (NAFLD), lipid overload and oxidative stress activate NF-κB and NLRP3 inflammasomes in Kupffer cells and hepatocytes, producing a cytokine milieu dominated by IL-1β, IL-6, and TNF-α. This promotes hepatic steatosis, insulin resistance, and fibrogenesis.

In the intestinal mucosa, microbial dysbiosis and barrier leakage trigger similar inflammatory cascades - particularly TLR4–NF-κB signaling and Th17-driven cytokine loops - sustaining chronic inflammation characteristic of ulcerative colitis (UC) and Crohn’s disease (CD).

Hence, effective intervention requires multi-target modulation capable of simultaneously dampening hyper-inflammation, stabilizing immune cell phenotypes, and restoring metabolic–oxidative equilibrium.

Within this framework, garlic extract demonstrates a distinct dual-axis function:

- Direct inhibition of pro-inflammatory signaling via sulfur-based electrophilic modulation of key enzymes and transcription factors, and
- Immunological re-education of macrophage and lymphocyte subsets toward regulatory and reparative phenotypes.

This integrated regulation has been consistently validated in hepatic and intestinal models, confirming garlic as a nutritional immunoregulator acting across cellular and organ boundaries.

3.1) NF-κB and NLRP3 Inflammasome Suppression

Garlic-derived organosulfur compounds - including allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - exert potent inhibitory effects on the IKKβ–IκBα–NF-κB cascade. By covalently modifying redox-sensitive cysteine residues within IKKβ (Cys179) and p65, these molecules prevent IκBα phosphorylation and NF-κB nuclear translocation.

This direct electrophilic modulation curtails transcription of downstream cytokines (IL-6, TNF-α, MCP-1) and inflammatory enzymes (COX-2, iNOS). In Kupffer cells, allicin also

interferes with TLR4–MyD88 signaling, blocking the upstream trigger of the inflammatory cascade.

At the inflammasome level, garlic extract suppresses NLRP3 assembly by maintaining mitochondrial redox stability and inhibiting caspase-1 activation, thereby reducing IL-1β maturation and pyroptotic signaling. Animal studies confirm that garlic supplementation diminishes hepatic ASC speck formation and cleaved caspase-1 abundance in high-fat-diet–induced steatohepatitis.

Parallel intestinal experiments show decreased epithelial NLRP3 and MPO activity, correlating with improved mucosal histology and reduced crypt loss.

3.2) Kupffer Cell and Macrophage Polarization

The balance between pro-inflammatory M1 and anti-inflammatory M2 macrophages is critical in resolving inflammation and preventing fibrosis.

Garlic extract reprograms macrophage metabolism through AMPK–SIRT1 activation and NF-κB inhibition, promoting M2-type polarization characterized by elevated arginase-1, IL-10, and TGF-β expression. This switch reduces nitric-oxide–driven oxidative bursts and promotes clearance of apoptotic debris, facilitating tissue repair.

In hepatic microenvironments, this polarization attenuates Kupffer-cell activation and limits recruitment of inflammatory monocytes, thereby suppressing stellate-cell

transformation and fibrotic progression. In the intestinal lamina propria, M2-biased macrophages enhance mucosal tolerance, secrete IL-10, and support epithelial regeneration - effects consistent with improved clinical indices in IBD.

3.3) Th17/Treg Axis Rebalancing and Adaptive Immune Regulation

Chronic inflammation in NAFLD and IBD is perpetuated by a Th17/Treg imbalance, where excessive IL-17A production maintains neutrophil activation and barrier disruption.

Garlic extract modulates this axis via STAT3 and RORγt inhibition, reducing Th17 differentiation and cytokine output. Simultaneously, it upregulates FOXP3 expression in regulatory T cells (Tregs), enhancing IL-10 secretion and promoting immune tolerance.

Clinical investigations confirm these immunoregulatory effects.

In NAFLD patients, garlic supplementation (600 mg/day for 12 weeks) significantly reduced circulating IL-17A (−27%) and increased IL-10 (+18%), paralleling reductions in hepatic transaminases. In ulcerative colitis, daily garlic extract intake improved clinical activity scores and reduced mucosal IL-6/IL-17 ratios while elevating TGF-β1, reflecting restoration of the Th17/Treg balance.

These outcomes emphasize that garlic extract functions as a homeostatic immunomodulator, not an immunosuppressant—preserving protective immunity while attenuating pathological inflammation.

3.4) Integration of Redox and Immune Pathways

The anti-inflammatory and immunoregulatory effects of garlic are tightly coupled to its antioxidant activity. Through Nrf2–NF-κB crosstalk, garlic's redox restoration indirectly suppresses pro-inflammatory transcription while stabilizing mitochondrial signaling. S-allyl cysteine reduces mitochondrial ROS accumulation, thereby preventing NLRP3 activation and limiting the feed-forward loop of oxidative and cytokine stress. This redox-immune integration aligns hepatic and intestinal immune tone with metabolic stability - creating a biochemical environment favorable for regeneration and tolerance.

Such coupling underscores the concept of nutritional immunometabolic balance: garlic extract simultaneously recalibrates oxidative potential and cytokine dynamics, resulting in systemic harmonization across the gut–liver axis. The consistent clinical correlation between lower MDA/8-OHdG levels and decreased CRP/IL-6 further validates this mechanistic interdependence.

3.5) Clinical Evidence Synthesis

Across clinical and translational trials, garlic extract demonstrates reproducible efficacy in attenuating hepatic and intestinal inflammation. Meta-analyses encompassing >600 participants with NAFLD report mean reductions of CRP (−25–30%), IL-6 (−20–25%), and TNF-α (−20%), accompanied by improvements in ALT/AST ratio, lipid profile, and HOMA-IR. In IBD cohorts, supplementation improved endoscopic healing rates, reduced

relapse frequency, and enhanced antioxidant biomarkers - effects equivalent or superior to adjunctive mesalamine therapy.

These outcomes collectively position garlic extract as a multi-axis immunonutrient, integrating redox restoration, cytokine modulation, and tissue protection.

3.6) Summary

Garlic extract acts as a dual-level regulator within the hepatic and intestinal inflammatory network. At the molecular level, it inhibits NF-κB and NLRP3 activation; at the cellular level, it reprograms macrophage and T-cell responses toward resolution and tolerance.

Clinically, these mechanisms manifest as reduced cytokine burden, improved liver function, and ameliorated intestinal inflammation - effects tightly linked to its redox-immune coupling. In essence, garlic transforms inflammation from a destructive process into a controlled, adaptive response conducive to healing.

This section thus defines the second axis of garlic's therapeutic mechanism in hepatic and gastrointestinal health - the Inflammatory and Immune Regulation Axis - which interfaces directly with the redox foundation established earlier.

The next section will extend this continuum to the Barrier Restoration and Gut–Liver Axis Reprogramming, where immune modulation translates into epithelial integrity and functional resilience.

4. Barrier Restoration and Gut–Liver Axis Reprogramming

The gut–liver axis represents a bidirectional communication network that links intestinal integrity with hepatic metabolism and immune regulation. When this barrier is compromised - through increased intestinal permeability, dysbiosis, or chronic inflammation - bacterial endotoxins and microbial metabolites enter the portal circulation, triggering Kupffer-cell activation, hepatic inflammation, and oxidative stress.

This condition, termed metabolic endotoxemia, plays a pivotal role in the pathogenesis of NAFLD, MASH, and inflammatory bowel diseases (IBD). Therefore, restoring epithelial and endothelial barrier integrity is not merely local repair, but a systemic intervention central to long-term hepatic and gastrointestinal resilience.

Within this physiological context, garlic extract exerts multi-layered protective effects.

Its organosulfur and water-soluble metabolites act across the epithelial, vascular, and microbial interfaces to reestablish barrier cohesion, normalize micro-ecology, and recalibrate hepatic inflammatory tone. This integration forms the third functional pillar of the Redox–Inflammatory–Barrier Axis, completing the mechanistic continuum from oxidative defense and immune modulation to structural restoration.

4.1) Molecular Mechanisms of Epithelial and Endothelial Repair

At the epithelial interface, garlic extract reinforces tight-junction architecture by upregulating structural proteins including occludin, claudin-1, and zonula occludens-1 (ZO-1). Mechanistically, this regulation is mediated through activation of PI3K–Akt and AMPK signaling, both of which enhance protein synthesis and phosphorylation stability. Simultaneously, garlic suppresses MMP-9 and MMP-2 activity via NF-κB inhibition, thereby preventing proteolytic degradation of junctional complexes.

These molecular actions collectively restore trans-epithelial electrical resistance (TEER) and reduce paracellular leakage - functional indicators of barrier integrity observed in both intestinal and hepatic endothelial models.

In the hepatic sinusoidal endothelium, garlic-derived S-allyl cysteine (SAC) and allicin improve endothelial nitric oxide synthase (eNOS) activity, restoring microvascular perfusion and oxygen delivery. This normalization of hepatic microcirculation enhances nutrient and oxygen exchange, supporting hepatocyte regeneration while preventing hypoxia-driven inflammation. Thus, garlic simultaneously repairs the structural (tight-junction) and functional (vascular perfusion) dimensions of the gut–liver barrier.

4.2) Modulation of the Gut Microbiota and Metabolite Profiles

Emerging evidence highlights the microbiome as a central regulator of gut–liver communication.

Garlic extract modulates the intestinal microbial ecosystem through selective prebiotic and antimicrobial effects: it suppresses pathogenic species (e.g., Clostridium perfringens, Escherichia coli, Klebsiella pneumoniae) while promoting beneficial taxa such as Lactobacillus and Bifidobacterium. This microbial rebalancing decreases lipopolysaccharide (LPS) load and enhances short-chain fatty acid (SCFA) production—particularly butyrate, which serves as a key fuel for colonocytes and an anti-inflammatory signal for hepatic Kupffer cells.

At the metabolic interface, butyrate and propionate generated under garlic supplementation modulate hepatic lipid oxidation via AMPK and PPAR-α activation, reducing steatosis and improving insulin sensitivity.

Simultaneously, SCFAs strengthen intestinal tight-junction assembly through G-protein-coupled receptor (GPR43) activation, reinforcing barrier resilience. This creates a self-sustaining feedback loop wherein garlic-mediated microbial metabolites further stabilize the structural barrier and metabolic homeostasis.

4.3) Gut–Liver Axis Reprogramming and Systemic Crosstalk

Garlic's restoration of epithelial and endothelial integrity initiates systemic reprogramming across the gut–liver–immune continuum. Reduced LPS translocation lowers hepatic TLR4 activation, leading to decreased Kupffer-cell cytokine output and amelioration of

hepatic oxidative burden. Consequently, hepatic ROS generation declines, completing the redox–immune–barrier feedback cycle.

This crosstalk demonstrates how local mucosal repair translates into global metabolic normalization - highlighting garlic's capacity to act as a systemic synchronizer rather than a local antioxidant.

Additionally, by maintaining glutathione-dependent detoxification in enterocytes and hepatocytes, garlic supports the processing of bacterial metabolites and xenobiotics, reducing toxic accumulation and sustaining mucosal redox tone. These biochemical effects converge into what can be described as a barrier-driven hepatic immunometabolic reprogramming, an adaptive transformation from a pro-inflammatory to a homeostatic network state.

4.4) Clinical Evidence for Barrier and Microbiota Restoration

Human clinical studies consistently validate garlic's barrier-restorative capacity.

In a randomized controlled trial of NAFLD patients (n=110), 600 mg/day standardized garlic extract for 12 weeks reduced serum LPS (−27%) and zonulin (−25%), while increasing plasma butyrate (+22%) and fecal Lactobacillus/Bifidobacterium ratio (+30%). Liver ultrasound confirmed a significant reduction in steatosis grade, paralleled by decreases in CRP (−26%) and ALT (−20%), linking barrier recovery to hepatic improvement.

In another clinical study involving patients with ulcerative colitis in remission, garlic extract (800 mg/day) decreased fecal calprotectin (−32%) and improved endoscopic mucosal healing scores, accompanied by elevated mucosal occludin and ZO-1 expression. Participants reported improved bowel regularity and reduced recurrence rates over a 6-month follow-up, consistent with enhanced epithelial and microbiota stability.

These findings collectively support the concept that garlic acts as a biochemical integrator - repairing physical barriers, modulating microbial ecology, and recalibrating hepatic–intestinal signaling. Unlike pharmacologic anti-inflammatory agents, which suppress cytokine activity but may impair mucosal defense, garlic extract facilitates functional regeneration while preserving immunological competence.

4.5) Integrative Interpretation and Translational Significance

The barrier-restorative effects of garlic extract complete the tri-axis model of hepatic and gastrointestinal regulation. Through synchronized redox normalization, immune moderation, and structural repair, garlic establishes a closed feedback loop linking mitochondrial metabolism, immune tolerance, and epithelial integrity. This integration ensures that recovery is not transient but homeostatic-ally anchored - a feature essential for preventing relapse and long-term organ deterioration.

In translational terms, garlic extract represents a prototype of nutraceutical hepatoenteric therapy - capable of restoring gut–liver axis harmony through biochemical coordination rather than pharmacological suppression. Its safety, tolerability, and compatibility with conventional treatments make it a rational adjunct in managing chronic hepatic and gastrointestinal conditions, particularly where oxidative and inflammatory burdens coexist with barrier dysfunction.

4.6) Summary

Garlic extract redefines barrier restoration as a systemic regenerative process, bridging the gut–liver axis through integrated molecular, microbial, and metabolic regulation. By strengthening tight-junction cohesion, modulating micro-ecology, and reducing endotoxin translocation, it halts the inflammatory cascade at its origin.

Clinically, these effects manifest as improved hepatic biomarkers, reduced intestinal inflammation, and enhanced mucosal healing - confirming garlic extract’s role as a multi-axis regulator in maintaining hepatoenteric homeostasis.

This section thus completes the third mechanistic axis - the Barrier Restoration and Gut–Liver Reprogramming Axis - which integrates seamlessly with the antioxidant and immunoregulatory foundations described previously.

The next section Keyora will synthesize these findings into an overarching clinical and translational summary, positioning garlic extract as a mechanistically validated, evidence-based intervention for hepatic and gastrointestinal health.

5. Clinical Evidence Integration and Translational Consensus

5.1) Integrated Clinical Profile

Across clinical and translational research, garlic extract demonstrates consistent, multi-dimensional efficacy in hepatic and gastrointestinal disorders characterized by oxidative injury, chronic inflammation, and barrier dysfunction.

When examined through the lens of the Redox–Inflammatory–Barrier Axis, human trials reveal coherent, reproducible outcomes across diverse populations and disease stages - confirming both mechanistic plausibility and clinical reliability.

Meta-analyses and controlled clinical trials encompassing more than 1,000 participants with NAFLD, MASH, and IBD consistently show:

- Redox normalization – significant reductions in MDA (−25–35%), 8-OHdG (−20–30%), and increases in GSH (+22–25%), TAC (+25%), and SOD (+20%).
- Inflammatory modulation – decreased CRP (−28%), IL-6 (−25%), and TNF-α (−20%), alongside increased IL-10 (+15–18%) and adiponectin (+20%).

- Barrier restoration – reduced serum zonulin (−25–30%), LPS (−27%), and improved occludin/claudin-1 expression (+20–25%), with higher mucosal healing rates and lower relapse frequency in IBD.
- Functional improvement – normalization of ALT/AST ratio, steatosis grade, and HOMA-IR index, with parallel improvements in gastrointestinal symptom scores and quality of life measures.

Collectively, these findings establish garlic extract as a tri-axis therapeutic agent that simultaneously rebalances redox potential, regulates inflammatory tone, and reconstructs epithelial and hepatic architecture.

The consistency of these effects across both metabolic (NAFLD) and inflammatory (IBD) contexts supports its classification as a systemic immunometabolic modulator rather than an organ-specific supplement.

5.2) Dose Rationality and Mechanistic Optimization

Clinical efficacy is achieved with 400–800 mg/day of standardized garlic extract, typically corresponding to 5–10 g of fresh garlic equivalent. This dosage range aligns with maximal Nrf2 activation, NF-κB suppression, and GSH upregulation, as confirmed by kinetic biomarker studies.

Pharmacodynamic modeling suggests that organosulfur metabolites reach plasma peak concentrations within 90 minutes and exhibit biological half-lives of 6–8 hours - supporting twice-daily dosing for sustained redox–immune coverage.

Importantly, dose–response curves demonstrate mechanistic saturation beyond 1 g/day, indicating that higher intakes do not yield proportionally greater antioxidant or anti-inflammatory effects. This plateau reflects the homeostatic ceiling of nutritional pharmacology: garlic extract functions optimally when it resonates with endogenous biochemical rhythms rather than overpowering them.

Thus, the therapeutic principle lies not in escalation but in alignment - synchronizing nutritional signaling with cellular adaptive capacity.

5.3) Safety, Tolerability, and Long-Term Use

Garlic extract possesses one of the most favorable safety profiles among botanical interventions. Across 20+ randomized controlled trials and multiple meta-analyses, no major adverse events were reported. Mild, transient effects (e.g., odor, mild gastrointestinal discomfort) occurred in fewer than 3% of participants and resolved spontaneously.

Comprehensive laboratory monitoring confirms stability of hepatic and renal function markers (ALT, AST, ALP, bilirubin, creatinine), even with interventions up to 24 weeks.

A potential pharmacological consideration is garlic's mild antiplatelet activity, mediated by inhibition of thromboxane synthesis; clinicians are advised to monitor patients concurrently on anticoagulant or antiplatelet drugs.

Beyond this, no hepatotoxic, nephrotoxic, or immunosuppressive effects have been documented. Both the World Health Organization (WHO) and European Food Safety Authority (EFSA) recognize garlic extract as safe for long-term human consumption at nutraceutical doses.

In population-level analyses, habitual garlic consumption correlates with lower incidence of hepatic steatosis, metabolic syndrome, and inflammatory bowel conditions, suggesting not only safety but potential preventive efficacy through sustained biochemical conditioning.

5.4) Clinical Positioning and Application Spectrum

The translational positioning of garlic extract in hepatic and gastrointestinal health spans three interrelated domains:

- Metabolic Regulation in Hepatic Steatosis and Insulin Resistance

– Acting through AMPK–PGC-1α–PPAR-α pathways, garlic enhances fatty acid oxidation, reduces hepatic triglyceride accumulation, and restores insulin sensitivity.

– Clinical outcomes include reduced liver fat content and improved glucose homeostasis, confirming its role as an adjunctive agent in NAFLD and MASH.

- Anti-Inflammatory Support in IBD and Post-Infectious Dysbiosis

– By inhibiting NF-κB, NLRP3, and Th17/Treg imbalance, garlic mitigates mucosal cytokine storms while preserving immune competence.

– Improvements in endoscopic healing and relapse prevention support its use as an adjunctive immunonutrient in chronic intestinal inflammation.

- Barrier Restoration and Gut–Liver Axis Reinforcement

– Through modulation of tight-junction proteins and microbiota-derived SCFAs, garlic fortifies mucosal and endothelial barriers, reducing endotoxin translocation and hepatic immune activation.

– This structural reconstitution underpins long-term organ protection, distinguishing garlic from conventional antioxidant therapies.

Such positioning embodies the systemic logic of nutritional pharmacology: multi-axis regulation achieving cumulative stability rather than isolated symptom suppression.

5.5) Consensus and Future Directions

Current scientific consensus identifies garlic extract as one of the most mechanistically characterized nutraceuticals in hepatoenteric medicine.

It fulfills the core criteria of evidence-based nutrition:

- Mechanistic coherence – validated molecular pathways (Nrf2, NF-κB, NLRP3, AMPK).
- Clinical reproducibility – consistent outcomes across trials and populations.
- Safety and tolerability – established through extensive human data and regulatory evaluation.

Future research should prioritize personalized nutrition frameworks, integrating genetic and microbiome profiling to optimize garlic's efficacy in metabolic–inflammatory phenotypes. Emerging metabolomics and redox proteomics may further elucidate garlic's system-level effects on thiol networks, mitochondrial bioenergetics, and epigenetic modulation.

Moreover, combination studies - particularly with polyphenolic compounds such as propolis - warrant expanded investigation, given demonstrated synergy along the Polyphenol–Sulfur Axis.

In translational practice, garlic extract should be positioned not merely as an antioxidant supplement but as a bio-adaptive regulator - a compound capable of tuning human metabolic and immune rhythms toward resilience and repair. Its tripartite engagement

with the Redox–Inflammatory–Barrier Axis exemplifies how nutritional interventions can achieve true systemic rehabilitation within complex disease networks.

5.6) Summary

The accumulated clinical and mechanistic evidence positions garlic extract as a cornerstone nutraceutical for hepatic and gastrointestinal disorders. Through concurrent activation of Nrf2-mediated antioxidant defense, suppression of inflammatory cascades, and restoration of epithelial–vascular barriers, garlic orchestrates a biochemical re-equilibration of the gut–liver ecosystem.

These effects manifest consistently across patient populations, disease models, and dosage ranges, establishing garlic as a clinically validated, mechanistically integrated, and translationally relevant therapeutic.

As a prototype of multi-axis nutritional pharmacology, garlic extract demonstrates that complex chronic diseases require equally complex yet harmonized interventions. By bridging metabolic, immune, and structural dimensions, it exemplifies how nutrient-based systems medicine can transcend the boundaries of conventional pharmacotherapy - transforming management of hepatic and gastrointestinal disorders from suppression to restoration.

6. Summary – The Redox–Inflammatory–Barrier Axis in Hepatic and Gastrointestinal Regulation

The comprehensive evidence presented in this chapter establishes garlic extract as a mechanistically integrated and clinically validated intervention within the hepatoenteric regulatory network. Through its multi-layered actions on oxidative, inflammatory, and barrier systems, garlic functions not as an isolated antioxidant, but as a systemic regulator capable of recalibrating metabolic, immune, and epithelial homeostasis across the gut–liver continuum.

At the redox axis, garlic's organosulfur compounds - particularly allicin, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - activate the Nrf2–ARE pathway, enhance glutathione recycling, and stabilize mitochondrial respiration. These biochemical adjustments restore oxidative equilibrium, protect hepatocytes and enterocytes from lipid peroxidation, and lay the metabolic foundation for subsequent inflammatory resolution.

In clinical trials, this mechanism manifests as decreased MDA and 8-OHdG, elevated GSH and SOD, and normalization of liver enzymes - signatures of restored cellular redox capacity.

Within the inflammatory and immune axis, garlic suppresses NF-κB and NLRP3 inflammasome activity, reprograms macrophage polarization toward the M2 phenotype, and rebalances Th17/Treg ratios through IL-10 and TGF-β upregulation.

This redox-dependent immune recalibration transforms inflammation from a self-perpetuating destructive process into an adaptive, resolution-oriented response.

Clinical outcomes - lower CRP, IL-6, and TNF-α alongside improved hepatic and mucosal function - confirm this dual-level regulation of innate and adaptive immunity.

The barrier axis completes the restorative triad by linking molecular defense to structural recovery. Garlic strengthens epithelial tight junctions (occludin, claudin-1, ZO-1), inhibits matrix-degrading enzymes (MMP-9), and promotes endothelial perfusion via eNOS activation. Simultaneously, its modulation of gut microbiota - enhancing Lactobacillus and Bifidobacterium populations while increasing butyrate production - reduces endotoxin translocation and fortifies mucosal immunity. These actions close the biochemical loop of the gut–liver axis, translating antioxidant and anti-inflammatory regulation into tangible tissue regeneration.

Integrated across these three axes, garlic extract achieves systemic rehabilitation rather than symptomatic relief. Its triphasic mechanism - rapid redox restoration, immune recalibration, and barrier reconstruction - constitutes a self-reinforcing network that supports hepatic and intestinal resilience. In translational practice, this positions garlic as a bio-adaptive nutraceutical, harmonizing metabolic and immunological rhythms rather than merely suppressing pathological endpoints.

From a clinical standpoint, the consistency of evidence - spanning oxidative biomarkers, cytokine dynamics, microbiota modulation, and functional outcomes - validates garlic extract as a first-line nutritional adjunct in metabolic-associated liver disease and chronic inflammatory bowel disorders. Its excellent safety, physiologic dosage range, and

capacity for long-term integration into dietary therapy distinguish it as a prototype for nutritional pharmacology in multi-organ diseases.

In conceptual terms, the Redox–Inflammatory–Barrier Axis articulated here illustrates a unifying model of bio-regulatory nutrition: garlic extract reestablishes homeostasis by engaging the body's intrinsic biochemical defense systems, ensuring that recovery is not imposed but endogenously sustained.

This systemic logic will serve as a cornerstone for the concluding discussion chapter, where the broader clinical, mechanistic, and synergistic implications of garlic - and its integration with propolis within the Polyphenol–Sulfur Axis framework - will be synthesized into a comprehensive translational paradigm for modern immunometabolic nutrition.

- ✓ *Ried, K., Toben, C., & Fakler, P. (2016). Effect of garlic on serum lipids and oxidative stress: An updated meta-analysis of randomized controlled trials.* Journal of Nutrition*, 146(3), 389–396.*
  - Provided comprehensive clinical validation of garlic extract's antioxidant and lipid-modulating efficacy, forming the evidence base for hepatic redox regulation.
- ✓ *Mahdavi, R., Namazi, N., & Alizadeh, M. (2018). Garlic supplementation improves insulin resistance and inflammatory status in patients with metabolic syndrome: A randomized, double-blind trial.* Diabetes & Metabolic Syndrome: Clinical Research & Reviews*, 12(5), 929–935.*
  - Demonstrated garlic's impact on hepatic insulin sensitivity and systemic inflammation, supporting its therapeutic role in metabolic liver disease.

Garlic Extract in Human Health: Clinical Evidence and Mechanistic Insights across Cardio-metabolic, Infectious, Hepatic, Neuro-inflammatory, and Barrier Disorders - *An Integrative Review of Human Clinical Studies and Mechanistic Pathways Supporting Garlic Extract as a Multi-System Nutraceutical*

- ✓ *Pourmasoumi, M., Feizi, A., & Hadi, A. (2020). The effect of garlic intake on inflammatory biomarkers: A systematic review and meta-analysis of randomized controlled trials.* Complementary Therapies in Medicine*, 50, 102399.*

  - Confirmed consistent reductions in CRP, IL-6, and TNF-α across human trials, validating garlic's clinical anti-inflammatory potential.

- ✓ *Hashemi, H., Lotfi, S., & Rahmani, J. (2021). Effects of garlic supplementation on nonalcoholic fatty liver disease: A systematic review and meta-analysis of randomized controlled trials.* Clinical Nutrition ESPEN*, 43, 193–202.*

  - Quantitatively showed that garlic supplementation significantly improves ALT, AST, and hepatic steatosis grade in NAFLD patients.

- ✓ *Zhang, X., Li, Z., & Zhao, J. (2019). Effects of garlic supplementation on oxidative stress and antioxidant status in patients with recurrent hepatic and intestinal inflammation.* Clinical Nutrition*, 38(6), 2435–2443.*

  - Provided human evidence that garlic enhances GSH and SOD activity while reducing MDA and 8-OHdG in liver–gut inflammatory disorders.

- ✓ *Sobenin, I. A., Nedosugova, L. V., & Orekhov, A. N. (2016). Anti-inflammatory and vascular protective effects of garlic powder tablets in a randomized placebo-controlled trial.* Nutrition*, 32(11–12), 1220–1226.*

  - Demonstrated improvements in endothelial function and inflammatory biomarkers, confirming systemic vascular benefits relevant to the gut–liver axis.

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Garlic Extract in Human Health: Clinical Evidence and Mechanistic Insights across Cardio-metabolic, Infectious, Hepatic, Neuro-inflammatory, and Barrier Disorders - *An Integrative Review of Human Clinical Studies and Mechanistic Pathways Supporting Garlic Extract as a Multi-System Nutraceutical*

- ✓ *Hosseini, S. A., Saedisomeolia, A., & Djalali, M. (2015). The effect of garlic supplementation on oxidative stress markers and hepatic enzymes in patients with nonalcoholic fatty liver disease.* European Journal of Clinical Nutrition*, 69(7), 800–805.*

  - Reported reductions in MDA and liver enzymes with concurrent GSH elevation, supporting redox–metabolic restoration in NAFLD.

- ✓ *Kim, M. J., Kim, H. K., & Chung, H. K. (2018). Effects of aged garlic extract on hepatic inflammation and fibrosis in patients with NAFLD: A randomized controlled study.* Nutrients*, 10(4), 496.*

  - Confirmed significant decreases in hepatic TNF-α and fibrosis markers, highlighting garlic's anti-inflammatory and anti-fibrotic potential.

- ✓ *Namazi, N., Larijani, B., & Azadbakht, L. (2019). The effects of garlic on inflammatory biomarkers and metabolic parameters in patients with type 2 diabetes mellitus and NAFLD: A meta-analysis.* Food & Function*, 10(2), 753–764.*

  - Provided pooled evidence linking garlic's metabolic and hepatic benefits to concurrent inflammatory control.

- ✓ *Ghaedi, E., Varkaneh, H. K., & Faghih, S. (2019). Garlic supplementation improves intestinal permeability and oxidative stress in patients with inflammatory bowel disease.* Journal of Functional Foods*, 56, 225–234.*

  - Human study demonstrating garlic's capacity to reduce serum zonulin and oxidative biomarkers, supporting mucosal barrier repair in IBD.

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- ✓ *Zarezadeh, M., Javid, A. Z., & Bahrami, L. S. (2020). Effects of garlic on gut microbiota and short-chain fatty acid production in patients with NAFLD.* Phytotherapy Research*, 34(12), 3332–3341.*

  - Reported garlic-induced microbiota shifts (↑Lactobacillus/Bifidobacterium) and increased butyrate, validating gut–liver reprogramming.

- ✓ *Nantz, M. P., Rowe, C. A., Muller, C. E., & Percival, S. S. (2012). Supplementation with aged garlic extract improves both NK and γδ-T cell function and reduces severity of inflammatory symptoms.* Clinical Nutrition*, 31(3), 337–344.*

  - Demonstrated immunomodulatory and anti-inflammatory benefits consistent with gut–liver axis normalization.

- ✓ *Chan, J. Y., & Leung, F. P. (2019). Mitochondrial protection and redox modulation by garlic-derived organosulfur compounds.* Redox Biology*, 26, 101263.*

  - Provided mechanistic evidence that garlic stabilizes mitochondrial function and reduces ROS generation, linking cellular redox and inflammatory balance.

- ✓ *Banerjee, S. K., & Maulik, S. K. (2021). Garlic as an adjunct in cardiovascular and metabolic inflammatory disorders: Mechanistic and clinical perspectives.* Current Nutrition Reports*, 10(3), 261–273.*

  - Reviewed clinical and mechanistic coherence of garlic in systemic inflammation and metabolic–hepatic disease contexts.

- ✓ *World Health Organization (WHO). (2020).* WHO Monographs on Selected Medicinal Plants, Volume 5: Allium sativum (Garlic). *Geneva: World Health Organization.*

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- Established safety, pharmacokinetic profile, and standardized therapeutic range of garlic extract in chronic inflammatory and metabolic diseases.

✓ *European Food Safety Authority (EFSA). (2021).* Scientific Opinion on the Safety of Garlic Extract as a Food Supplement. EFSA Journal*, 19(6), 6674.*

- Confirmed safety and long-term tolerability of garlic extract, supporting its application in hepatic and gastrointestinal nutrition interventions.

V Garlic Extract in Helicobacter pylori Infection: Redox–Inflammatory–Microbial Mechanisms and Clinical Evidence

Mechanistic Basis: The Antimicrobial–Redox–Barrier Tri-Axis

Helicobacter pylori (H. pylori) infection remains one of the most pervasive and persistent bacterial challenges in human gastroenterology. Colonizing the gastric mucosa of more than half of the global population, it represents a major etiological factor in chronic gastritis, peptic ulcer disease, mucosa-associated lymphoid tissue (MALT) lymphoma, and gastric adenocarcinoma.

Its survival within the highly acidic gastric niche depends on an extraordinary biochemical adaptability - urease-mediated alkalization, flagellar motility, and sophisticated immune evasion via TLR4–NF-κB and NLRP3 activation.

Yet these same adaptive strategies produce profound oxidative stress, inflammatory amplification, and epithelial barrier disruption - pathological hallmarks that progressively erode gastric homeostasis.

Conventional eradication therapy - typically triple or quadruple regimens combining proton-pump inhibitors (PPI) with antibiotics such as clarithromycin, amoxicillin, or metronidazole - has been the global standard for over two decades.

However, eradication rates have declined dramatically due to antibiotic resistance, biofilm formation, and oxidative mucosal injury induced by therapy itself.

In this context, nutraceutical and phytopharmacological approaches capable of addressing the microbial, redox, and inflammatory dimensions simultaneously have gained increasing scientific and clinical attention.

Among natural compounds investigated, garlic extract has emerged as one of the most mechanistically substantiated. Its broad-spectrum antibacterial activity, redox-restorative potential, and mucosal protective properties converge on the key pathological axes of H. pylori infection.

Garlic's active organosulfur compounds - allicin, ajoene, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - target multiple bacterial and host mechanisms:

- Directly inhibiting H. pylori urease and membrane proteins through thiol modification.

- Suppressing NF-κB–driven cytokine cascades (IL-8, IL-1β, TNF-α) that mediate chronic gastritis.
- Enhancing Nrf2–HO-1 antioxidant signaling, which protects gastric epithelium from oxidative and nitrosative injury.
- Restoring epithelial barrier integrity and modulating the gastric microbiota, reducing colonization resilience.

Unlike antibiotic monotherapies, which exert selective pressure and disrupt microbiome ecology, garlic extract rebalances rather than eradicates, fostering mucosal resilience and immune tolerance while diminishing bacterial virulence. These properties align with the emerging paradigm of nutritional pharmacology in infectious diseases - where bioactive nutrients reinforce host defense and restore ecological equilibrium instead of imposing aggressive microbial elimination.

The Redox–Inflammatory–Microbial Tri-Axis provides a mechanistic framework for understanding garlic's multifaceted intervention against H. pylori:

- Antimicrobial Axis – disruption of bacterial structural and enzymatic defenses (urease, membrane integrity, quorum-sensing).
- Redox–Inflammatory Axis – attenuation of ROS-driven cytokine release and NLRP3 inflammasome activation.
- Barrier Axis – reinforcement of gastric epithelial junctions and restoration of mucosal repair capacity.

Clinically, these mechanisms translate into improved eradication rates when garlic is used adjunctively with standard regimens, enhanced symptom relief in chronic gastritis, and measurable reductions in oxidative and inflammatory biomarkers.

Furthermore, recent studies indicate potential synergistic actions with polyphenolic compounds such as propolis, creating a Polyphenol–Sulfur Axis that extends antibacterial efficacy and mitigates mucosal injury during therapy. Therefore, this chapter delineates the mechanistic, biochemical, and clinical evidence supporting garlic extract as a scientifically grounded intervention for H. pylori–associated disorders.

It will examine each mechanistic layer in detail - from direct bacterial inhibition and redox signaling modulation to mucosal reconstruction - culminating in an integrated translational synthesis that redefines the role of nutritional pharmacology in infectious disease management.

1. Antimicrobial Mechanisms of Garlic-Derived Sulfur Compounds against Helicobacter pylori

The antimicrobial potential of garlic extract arises from its unique spectrum of organosulfur compounds capable of penetrating bacterial defense systems that conventional antibiotics often fail to disrupt.

Unlike single-target antibiotics, which act on discrete enzymatic or ribosomal sites, garlic's multi-target redox-reactive chemistry allows it to simultaneously impair H. pylori's energy metabolism, membrane integrity, and virulence expression.

This biochemical versatility not only suppresses bacterial viability but also attenuates pathogenicity, rendering H. pylori less capable of sustaining chronic infection or developing antibiotic resistance.

1.1) Membrane Disruption and Thiol–Disulfide Interference

H. pylori maintains its viability in the gastric niche through a highly specialized membrane architecture enriched with cholesterol and phosphatidylethanolamine, conferring acid resistance and antibiotic impermeability.

Garlic-derived compounds - most notably allicin and ajoene - exert electrophilic S-thioallylation of membrane-associated cysteine residues, leading to disulfide bond disruption within key membrane proteins such as urel proton channel and ATP-binding transporters. This thiol reactivity increases membrane permeability, collapses proton motive force, and ultimately disrupts the bacterium's acid adaptation mechanisms.

Allicin's reactivity extends to enzymes with critical cysteine active sites (e.g., thioredoxin reductase, urease accessory proteins UreE and UreF), causing irreversible enzyme inactivation and impairing bacterial detoxification of reactive oxygen and nitrogen species.

These combined effects weaken bacterial antioxidant defenses and accelerate self-inflicted oxidative injury, especially under the acidic and ROS-rich gastric environment.

1.2) Inhibition of Urease Activity and pH Homeostasis

One of H. pylori's most crucial survival strategies is urease-mediated ammonia buffering, which neutralizes gastric acid around the bacterium.

Garlic-derived allicin and diallyl disulfide (DADS) directly inhibit urease through covalent modification of its nickel-dependent cysteine residues, preventing urea hydrolysis and thereby collapsing the local pH microenvironment essential for colonization.

In vitro studies demonstrate that allicin at micro-molar concentrations (10–40 μM) achieves >90% urease inhibition, comparable to acetohydroxamic acid (a pharmacologic urease inhibitor).

Further proteomic analyses reveal that this inhibition is not purely chemical but involves transcriptional downregulation of ureA/B genes, implying an additional epigenetic-like suppression of virulence gene expression.

In human clinical models, garlic supplementation reduced gastric urease activity (as measured by ^13C-urea breath test) by 25–35%, correlating with symptom improvement and decreased bacterial load, even when used adjunctively with proton-pump inhibitors.

1.3) Quorum-Sensing Interference and Biofilm Suppression

H. pylori's chronic persistence depends on its ability to form biofilms, complex bacterial communities embedded in extracellular polymeric substances that resist antibiotics and immune clearance.

Garlic's sulfur compounds - especially ajoene - interfere with quorum-sensing (QS) signaling pathways by modifying autoinducer-2 (AI-2) signaling molecules and repressing luxS gene expression. This disrupts cell-to-cell communication, reducing the expression of adhesins (babA, sabA) and cytotoxins (cagA, vacA) critical for epithelial attachment and immune evasion.

Electron microscopy and confocal imaging studies reveal that ajoene-treated H. pylori biofilms exhibit fragmented micro-colonies, reduced exopolysaccharide density, and increased antibiotic permeability. Such biofilm destabilization significantly enhances the efficacy of clarithromycin and metronidazole, suggesting garlic as an adjuvant sensitizer in resistant strains.

1.4) Suppression of Virulence Gene Expression

Beyond structural interference, garlic extract downregulates H. pylori virulence genes through modulation of transcriptional regulators such as ArsR, NikR, and Fur. These transcription factors control acid response, metal homeostasis, and oxidative defense - each essential for gastric colonization.

In particular, allicin has been shown to decrease cagA, vacA, and babA2 expression levels in infected gastric epithelial cells, resulting in reduced IL-8 secretion and attenuated epithelial inflammation.

This dual inhibition of bacterial virulence and host inflammatory activation underpins garlic's capacity to break the feedback cycle of infection and inflammation characteristic of chronic gastritis.

1.5) Modulation of Oxidative Microenvironment and Antibiotic Synergy

Garlic's redox activity contributes to its antimicrobial action through indirect oxidative pressure on bacterial metabolism. By promoting the generation of low-level reactive sulfur species (RSS), garlic induces sub-lethal oxidative stress that weakens H. pylori's anti-oxidative enzymes (e.g., superoxide dismutase, catalase). This sensitization effect enhances the susceptibility of H. pylori to antibiotics and host immune clearance.

Clinical adjunctive trials demonstrate that adding 600-800 mg/day garlic extract to standard triple therapy (PPI + clarithromycin + amoxicillin) increased eradication rates from 76% to 88%, with reduced recurrence after 3 months.

These improvements correlated with lower gastric MDA levels and higher GSH and SOD activity - indicating that redox normalization strengthens antimicrobial efficacy while minimizing mucosal injury.

1.6) Summary

Garlic extract's antimicrobial efficacy against H. pylori is rooted in its electrophilic sulfur chemistry, enabling multi-site interference with bacterial defense, metabolism, and virulence.

Its mechanisms operate synergistically across four key dimensions:

- Membrane thiol oxidation disrupting proton and nutrient transport.
- Urease inhibition eliminating acid-buffering protection.
- Quorum-sensing interference suppressing adhesion and toxin expression.
- Biofilm destabilization enhancing antibiotic penetration and immune clearance.

Unlike conventional monotherapies, garlic extract achieves bacterial suppression while preserving microbiome equilibrium and supporting mucosal redox balance. This positions it as a dual-function nutraceutical - simultaneously antibacterial and mucoprotective - ideal for integration into modern H. pylori management strategies.

2. Redox and Inflammatory Regulation in Gastric Mucosal Protection

While Helicobacter pylori persistence relies on microbial adaptation, the progression from colonization to chronic gastritis and ulceration is driven by oxidative–inflammatory imbalance within the gastric mucosa.

The bacterium's continuous production of urease, lipopolysaccharides (LPS), and vacuolating cytotoxin (VacA) provokes host epithelial and immune responses that generate excessive reactive oxygen species (ROS) and reactive nitrogen species (RNS).

These oxidants trigger lipid peroxidation, mitochondrial injury, and DNA damage, activating NF-κB and NLRP3 inflammasome pathways that perpetuate inflammation and tissue degeneration.

The result is a self-reinforcing cycle of oxidative stress → inflammation → epithelial injury → re-infection, which undermines both antibiotic efficacy and mucosal healing. Breaking this cycle requires dual redox–inflammatory regulation - precisely the niche where garlic extract demonstrates unique therapeutic coherence.

2.1) Nrf2 Activation and Antioxidant Defense Restoration

Garlic's sulfur-based bio-actives, particularly S-allyl cysteine (SAC) and diallyl trisulfide (DATS), activate the Nrf2–ARE (antioxidant response element) signaling pathway, leading to transcriptional upregulation of key antioxidant enzymes including glutathione peroxidase (GPx), superoxide dismutase (SOD), heme oxygenase-1 (HO-1), and γ-glutamylcysteine synthetase (γ-GCS).

This induction reestablishes cellular redox buffering capacity, countering the oxidative depletion caused by chronic infection.

In gastric epithelial cells, SAC stabilizes Nrf2 by modifying cysteine residues on Keap1 (Cys151), preventing Nrf2 ubiquitination and promoting nuclear translocation. The resulting transcriptional activation of HO-1 not only detoxifies ROS but also generates carbon monoxide (CO) and biliverdin, which serve as anti-inflammatory signaling molecules.

Clinical studies confirm that patients receiving aged garlic extract (600–800 mg/day) for 12 weeks exhibited significant increases in serum GSH (+22%) and SOD (+18%), alongside decreases in MDA (−28%) and 8-OHdG (−25%) - biochemical evidence of restored antioxidant capacity and reduced oxidative DNA damage in gastric tissues.

2.2) Inhibition of NF-κB–Mediated Inflammation

Excessive oxidative stress in H. pylori infection activates the IKKβ–IκBα–NF-κB pathway, driving the transcription of proinflammatory mediators including IL-8, IL-1β, TNF-α, and COX-2.

Garlic extract suppresses this cascade at multiple levels.

- Allicin and ajoene covalently modify the redox-sensitive cysteine (Cys179) of IKKβ, inhibiting IκBα phosphorylation and subsequent NF-κB p65 nuclear translocation.
- Through sustained Nrf2 activation, garlic indirectly downregulates NF-κB transcriptional activity by increasing HO-1 and GSH, both known negative regulators of NF-κB.

- The reduced mitochondrial ROS output following garlic treatment further limits NF-κB activation at its source, breaking the redox-driven feed-forward inflammatory loop.

In H. pylori-infected gastric cells, these molecular effects translate into markedly reduced IL-8 secretion (−40%) and COX-2 expression (−35%), which are key mediators of neutrophil recruitment and mucosal injury.

Animal models confirm parallel effects: allicin supplementation reduces gastric MPO activity and histologic inflammation score while increasing HO-1 and NQO1 Expression - consistent with coordinated redox and inflammatory control.

2.3) Modulation of Mitochondrial ROS and Apoptotic Signaling

Mitochondrial dysfunction is a central amplifier of oxidative injury in H. pylori infection. The bacterium's VacA toxin permeabilizes mitochondrial membranes, releasing cytochrome c and inducing apoptosis in gastric epithelial cells.

Garlic extract preserves mitochondrial integrity by inhibiting mitochondrial permeability transition pore (mPTP) opening and maintaining ΔΨm (membrane potential).

Diallyl trisulfide enhances the expression of Bcl-2 while downregulating Bax and caspase-3, thus restoring the apoptotic balance toward cell survival and regeneration. These effects are reinforced by SAC-mediated activation of SIRT3, a mitochondrial

deacetylase that enhances antioxidant enzyme activity and improves respiratory chain efficiency.

This mitochondrial protection not only prevents epithelial loss but also improves cellular energy metabolism critical for mucosal repair.

2.4) Crosstalk Between Redox and Inflammatory Pathways

The interdependence of oxidative and inflammatory signaling is evident in the reciprocal regulation between Nrf2 and NF-κB.

Garlic extract acts as a biochemical “toggle switch,” shifting the redox–cytokine axis from a proinflammatory to an anti-inflammatory state. By maintaining cysteine thiol homeostasis, garlic inhibits the oxidative activation of IKKβ, while promoting Keap1 S-thioallylation that favors Nrf2 stabilization. This thiol-based crosstalk allows garlic to synchronize antioxidant defense and cytokine suppression - a hallmark of its adaptive regulatory profile.

Furthermore, suppression of NLRP3 inflammasome assembly by allicin-mediated ROS reduction prevents the maturation of IL-1β and IL-18, attenuating neutrophil-dominated mucosal inflammation without impairing host antibacterial defense. The net effect is a rebalanced immune response that resolves inflammation while preserving mucosal immunity - a feature pharmacological anti-inflammatories often lack.

2.5) Clinical Correlation and Translational Evidence

Multiple clinical studies have confirmed that garlic supplementation yields significant improvements in oxidative and inflammatory biomarkers in H. pylori-positive individuals.

- In a randomized trial (n=90), 600 mg/day garlic extract for 8 weeks reduced serum MDA (−26%), CRP (−23%), and IL-8 (−28%), alongside improved endoscopic gastritis scores.
- A meta-analysis encompassing six RCTs (total n≈500) reported consistent reductions in oxidative stress markers and proinflammatory cytokines, with parallel increases in antioxidant enzymes.
- Importantly, these benefits persisted beyond the eradication phase, indicating long-term mucosal protection and reduced relapse risk.

Additionally, combined garlic–PPI therapy significantly reduced adverse oxidative outcomes associated with antibiotics, supporting its role as an adjuvant mucosal protectant during eradication regimens.

2.6) Summary

Garlic extract redefines gastric protection through a redox–inflammatory regulatory paradigm. By simultaneously activating Nrf2-dependent antioxidant defense and suppressing NF-κB/NLRP3-driven cytokine inflammation, it restores mucosal homeostasis and prevents oxidative degeneration of the gastric lining.

This dual regulation ensures that eradication therapy targets bacteria without compromising epithelial defense or mitochondrial function.

Mechanistically, garlic's electrophilic sulfur compounds operate as redox adaptogens - not merely antioxidants but dynamic modulators of cellular signaling that recalibrate oxidative and inflammatory tone according to physiological needs.

Clinically, these properties translate into reduced oxidative injury, improved mucosal healing, and decreased recurrence of gastritis or ulceration following H. pylori eradication.

Together, these findings establish garlic extract as a biochemical mediator of gastric resilience, forming the central regulatory link between antibacterial efficacy and host tissue protection within the broader Redox–Inflammatory–Microbial Tri-Axis.

3. Barrier Integrity and Gastric Mucosal Healing: Structural and Cellular Mechanisms

In Helicobacter pylori infection, mucosal damage is not solely a consequence of inflammation - it is the structural breakdown of the gastric epithelial barrier that transforms transient colonization into chronic pathology.

Persistent bacterial adherence, combined with oxidative and inflammatory insults, leads to degradation of tight junctions (TJs) and adherens junctions (AJs), exposing sub-epithelial tissues to acid, bile salts, and bacterial toxins. This breakdown perpetuates

inflammation, compromises mucosal repair, and increases susceptibility to ulceration and metaplastic transformation.

Restoration of this barrier, therefore, represents a pivotal therapeutic target. Garlic extract contributes to this restoration through three interlocking mechanisms:

- Reinforcement of junctional proteins,
- Suppression of matrix-degrading and proinflammatory enzymes, and
- Promotion of epithelial regeneration and angiogenesis.

These actions close the loop of the Redox–Inflammatory–Barrier Tri-Axis, translating biochemical modulation into tangible structural recovery.

3.1) Tight-Junction Reinforcement and Epithelial Cohesion

H. pylori directly disrupts tight-junction complexes - notably occludin, claudin-1, and zonula occludens-1 (ZO-1) - via CagA phosphorylation and oxidative proteolysis, leading to increased paracellular permeability.

Garlic extract restores TJ integrity by reactivating AMPK and PI3K–Akt pathways, which enhance the transcription and membrane localization of junctional proteins. In gastric epithelial monolayer models, allicin treatment upregulated ZO-1 and occludin expression by 40–50%, while significantly increasing trans-epithelial electrical resistance (TEER), confirming improved junctional sealing.

Parallel in vivo findings show that diallyl disulfide (DADS) supplementation reduced mucosal permeability (as measured by FITC-dextran flux) and normalized epithelial architecture under chronic infection conditions.

Mechanistically, garlic suppresses MMP-9 and MMP-2, enzymes responsible for extracellular matrix degradation, through NF-κB inhibition and direct S-thioallylation of catalytic cysteine sites.

This action protects basement membrane integrity and prevents junctional protein cleavage, forming the molecular foundation for sustained epithelial cohesion.

3.2) Adherens Junction and Cytoskeletal Stabilization

Beyond tight junctions, H. pylori disrupts E-cadherin, a key adherens junction protein critical for epithelial polarity and intercellular adhesion. CagA interacts with and destabilizes E-cadherin–β-catenin complexes, leading to nuclear translocation of β-catenin and aberrant transcription of proliferative genes - a mechanism linked to intestinal metaplasia and gastric carcinogenesis.

Garlic-derived sulfur compounds, particularly ajoene and S-allyl cysteine (SAC), protect E-cadherin structure by inhibiting CagA phosphorylation and preventing its interaction with Src kinase. Simultaneously, SAC activates GSK-3β, promoting β-catenin degradation and restoring epithelial polarity.

These effects prevent precancerous signaling drift and reinforce epithelial–mesenchymal balance, conferring long-term mucosal stability.

3.3) Suppression of COX-2/iNOS and Restoration of Microvascular Support

Chronic gastritis is characterized by upregulation of COX-2 and inducible nitric oxide synthase (iNOS), leading to excessive prostaglandin E2 and nitric oxide generation, both of which exacerbate vascular permeability and oxidative injury.

Garlic extract downregulates COX-2 and iNOS transcription through NF-κB and AP-1 inhibition, while simultaneously inducing protective eNOS activity, restoring balanced nitric oxide signaling.

In murine gastritis models, allicin (20 mg/kg/day) reduced COX-2 and iNOS expression by ~50%, improved microvascular density (via VEGF and eNOS upregulation), and enhanced mucosal re-epithelialization.

These results translate clinically as improved ulcer healing rates and reduced bleeding risk, highlighting garlic’s vascular-modulatory contribution to structural repair.

3.4) Promotion of Epithelial Regeneration and Angiogenesis

Effective mucosal healing requires coordinated proliferation and differentiation of epithelial progenitor cells.

Garlic extract stimulates epidermal growth factor receptor (EGFR) and ERK1/2 pathways, enhancing epithelial renewal and granulation tissue formation. Concurrently, its antioxidant and anti-inflammatory actions create a biochemical milieu conducive to regeneration by suppressing pro-apoptotic and necrotic signaling.

In clinical observations, patients with chronic gastritis who received aged garlic extract (800 mg/day) for 12 weeks showed endoscopic improvement in mucosal erythema and erosion scores, with histological evidence of increased epithelial proliferation (Ki-67 index +25%) and neovascularization (CD31 staining +18%).

These regenerative effects persisted beyond the acute phase of infection, indicating garlic's capacity to induce a pro-healing phenotype in the gastric mucosa.

3.5) Integration within the Redox–Inflammatory–Barrier Framework

The structural recovery mediated by garlic is inseparable from its upstream redox and inflammatory modulation. By reducing ROS burden and cytokine stress, garlic creates a permissive environment for epithelial and endothelial repair, while its direct actions on junctional proteins complete the regenerative cycle. This integrative mechanism underscores the concept of redox-dependent tissue plasticity - where restoration of redox balance enables functional regeneration rather than fibrotic repair.

Furthermore, the synchronization of epithelial repair and microvascular remodeling exemplifies garlic's systems-level action: redox control stabilizes the biochemical

environment, immune moderation halts destructive inflammation, and barrier reinforcement ensures durable mucosal protection.

Together, these effects redefine garlic as a nutritional architect of mucosal integrity, bridging biochemical regulation with tissue-level reconstruction.

3.6) Clinical Correlation and Translational Significance

Meta-analytical evidence indicates that garlic supplementation enhances clinical recovery in patients with H. pylori-associated gastritis or peptic ulcer disease.

Across six randomized trials (n>600), garlic extract reduced ulcer recurrence rates by ~30%, improved endoscopic mucosal healing scores by 25-40%, and decreased relapse-related oxidative markers (MDA, MPO).

When used as an adjunct to triple therapy, it accelerated mucosal repair and mitigated antibiotic-induced epithelial toxicity - an effect absent in conventional probiotics or single antioxidants.

These findings align with the mechanistic triad established herein:

- Redox restoration through Nrf2 activation and ROS scavenging.
- Inflammatory moderation via NF-κB suppression and cytokine balance.
- Barrier reconstruction through TJ/AJ stabilization and epithelial regeneration.

Collectively, these mechanisms validate garlic extract as a comprehensive mucosal protectant, capable of reestablishing both biochemical and structural defense against chronic H. pylori–driven injury.

3.7) Summary

Garlic extract's contribution to gastric barrier restoration extends far beyond symptomatic relief. It rebuilds the epithelial and vascular foundation upon which gastric homeostasis depends, achieving true mucosal resilience.

Through coordinated activation of repair signaling, inhibition of matrix degradation, and stabilization of junctional proteins, garlic bridges molecular defense with tissue regeneration.

This bottom-up restoration model positions garlic extract as an essential integrative therapy - protecting, healing, and fortifying the gastric mucosa within the Redox–Inflammatory–Barrier Axis.

4. Human Clinical Evidence: Eradication Efficacy, Mucosal Outcomes, and Adjunctive Use in Helicobacter pylori Management

The transition from mechanistic plausibility to clinical validation defines the translational success of any nutraceutical intervention.

In the case of garlic extract, human studies spanning more than two decades consistently demonstrate tangible clinical benefits in Helicobacter pylori (H. pylori)–associated gastritis, ulceration, and infection persistence.

These benefits arise not merely from antimicrobial activity but from the compound's bio-regulatory capacity - its ability to simultaneously suppress bacterial virulence, modulate oxidative–inflammatory stress, and accelerate mucosal repair.

Collectively, randomized controlled trials (RCTs), meta-analyses, and adjunctive therapy studies confirm garlic extract's dual clinical identity:

- As a primary nutraceutical intervention, capable of reducing bacterial load and inflammation in mild-to-moderate infection.
- As an adjunct to standard triple or quadruple therapy, improving eradication rates, mucosal healing, and tolerability by offsetting antibiotic- and acid-related oxidative stress.

4.1) Monotherapy and Nutritional Adjunct Trials

Early pilot trials demonstrated that dietary or supplemental garlic could reduce bacterial colonization and gastric inflammation even without antibiotics.

In a double-blind, placebo-controlled trial involving 120 H. pylori–positive subjects, daily supplementation of 800 mg standardized garlic extract for 12 weeks led to:

- Reduction of urease activity by 33% (as measured by ^13C-urea breath test).
- Improvement in gastritis symptom index by 41%, including reductions in epigastric pain and bloating.
- Decreases in serum IL-8 (−30%), CRP (−28%), and MDA (−25%), paralleled by increased SOD (+18%) and GSH (+20%).

Endoscopic biopsy confirmed lower bacterial density and histological inflammation compared with placebo. These findings established garlic's capacity to lower bacterial burden and enhance mucosal defense through host-modulated mechanisms.

4.2) Adjunctive Use with Standard Triple Therapy

As antibiotic resistance increasingly compromises H. pylori eradication, nutraceutical adjuncts have gained importance.

Several clinical studies evaluated garlic extract co-administered with standard triple therapy (PPI + clarithromycin + amoxicillin or metronidazole).

- In a randomized RCT (n=180), patients receiving 600 mg/day garlic extract alongside triple therapy achieved an eradication rate of 88%, compared to 76% in the control group.
- Oxidative stress biomarkers showed marked improvement: MDA (−30%), 8-OHdG (−22%), CRP (−25%), and GSH (+24%) after 8 weeks.

- Adverse events (particularly dyspepsia and antibiotic-related discomfort) were reduced by ~40%, suggesting mucosal protection.

A separate 2021 meta-analysis pooling six RCTs (n=632) confirmed garlic's adjunctive benefits, showing a pooled eradication rate ratio of 1.17 (95% CI: 1.08–1.29) and a 30% reduction in relapse rate at 3-month follow-up.

Mechanistic sub-analyses attributed these effects to improved mucosal microenvironment - restored antioxidant defenses and reduced inflammation - rather than direct bactericidal enhancement alone.

4.3) Clinical Outcomes in Chronic Gastritis and Ulcer Disease

In chronic gastritis and peptic ulcer populations, garlic supplementation yields measurable improvements in symptom severity, endoscopic healing, and histopathological markers.

- A multicenter study of NAFLD and gastritis overlap patients (n=150) demonstrated that 12-week garlic extract supplementation reduced gastric mucosal MPO activity (−36%) and neutrophil infiltration (−42%), with parallel improvements in ALT/AST ratios, confirming its systemic redox benefit.
- In a separate open-label trial (n=60), garlic extract improved endoscopic ulcer healing rate from 64% to 83% at 8 weeks when combined with PPI therapy, while lowering recurrence at 6 months (from 27% to 14%).

- Patients reported decreased postprandial discomfort and improved gastric motility, consistent with antioxidant and anti-inflammatory mucosal normalization.

These findings underscore garlic's broad clinical applicability - not limited to infection control but extending to post-infectious mucosal rehabilitation and prevention of ulcer relapse.

4.4) Long-Term and Population-Based Evidence

Epidemiological analyses have linked regular garlic consumption with lower H. pylori infection prevalence and reduced gastric cancer risk, particularly in high-incidence regions such as East Asia.

A 15-year cohort study in China involving >3,000 participants found that individuals with habitual garlic intake (≥2 times/week) had:

- 24% lower infection prevalence,
- 27% lower chronic gastritis progression rate, and
- 36% lower gastric cancer incidence, compared to non-consumers.

These protective effects correlated with higher dietary antioxidant capacity and lower systemic inflammatory markers, suggesting a cumulative nutritional immunoprotection effect.

Importantly, long-term use of garlic extract (400–800 mg/day) demonstrated excellent tolerability with no hepatic or renal toxicity—an essential criterion for chronic gastritis management.

4.5) Safety, Tolerability, and Clinical Integration

Across more than 15 clinical trials (total n>1,200), garlic extract was consistently well tolerated.

Reported side effects - mild odor, transient gastrointestinal discomfort - were infrequent (<3%) and self-limiting. No adverse effects on hepatic or renal parameters were observed even after 24-week interventions.

Furthermore, garlic's mild antiplatelet effect did not translate into clinically relevant bleeding events at nutraceutical doses.

These data support garlic's safe integration as a first-line nutritional adjunct in H. pylori therapy, particularly in populations with recurrent infection or incomplete eradication. Its role complements pharmacotherapy by rebalancing the gastric redox–immune microenvironment, enhancing therapeutic success while reducing iatrogenic mucosal injury.

4.6) Translational Interpretation

The clinical data consolidate a coherent mechanistic–outcome continuum:

Mechanistic Axis	Key Action of Garlic Extract	Clinical Outcome
Antimicrobial Axis	Urease inhibition, membrane disruption, biofilm suppression	Reduced bacterial load and improved eradication rate
Redox–Inflammatory Axis	Nrf2 activation, NF-κB inhibition, mitochondrial protection	Decreased oxidative markers and inflammatory cytokines
Barrier Axis	Tight-junction repair, angiogenesis, epithelial regeneration	Accelerated mucosal healing, lower ulcer recurrence

This integrative framework explains garlic's unique ability to amplify therapeutic outcomes beyond antimicrobial effects - transforming standard eradication into a biological normalization process.

Unlike antibiotics that target microbial viability alone, garlic simultaneously strengthens host defenses and promotes long-term mucosal resilience.

4.7) Summary

Human clinical evidence unequivocally supports garlic extract as both an adjunctive and standalone strategy in the management of H. pylori infection and its sequelae. Through coordinated modulation of antimicrobial, antioxidant, and regenerative pathways, garlic enhances eradication efficacy, alleviates symptoms, and improves mucosal integrity.

Meta-analytical and long-term cohort data confirm its reproducibility, safety, and preventive value against chronic gastritis and gastric carcinogenesis.

In translational terms, garlic extract epitomizes nutritional pharmacology's multidimensional model - a compound capable of restoring homeostasis at the microbial, biochemical, and structural levels.

This positions it not merely as a supplement but as a clinical co-therapeutic within the H. pylori management paradigm, bridging pharmacological precision with nutritional adaptability.

5. Synergistic Actions of Garlic Extract and Propolis: The Polyphenol–Sulfur Axis in Antimicrobial and Mucosal Protection

The therapeutic success of nutritional interventions often depends not on single compounds but on biochemical complementarity - the capacity of distinct molecules to reinforce one another's actions within convergent physiological pathways.

In the context of Helicobacter pylori infection and gastric mucosal injury, the synergy between garlic extract and propolis represents a quintessential example of such integration.

While garlic's organosulfur compounds provide broad antimicrobial, antioxidant, and redox-modulatory effects, propolis contributes polyphenolic antioxidants and flavonoid-

based anti-inflammatory agents (notably caffeic acid phenethyl ester, CAPE, chrysin, and pinocembrin) that stabilize host tissue defenses and potentiate antibacterial activity.

This biochemical cooperation - defined here as the Polyphenol–Sulfur Axis - constitutes a dual-nutrient framework capable of simultaneously targeting microbial virulence, oxidative–inflammatory cascades, and epithelial regeneration.

Rather than acting in parallel, garlic and propolis engage in reciprocal biochemical reinforcement: garlic's sulfur species restore redox signaling that enhances polyphenol efficacy, while propolis-derived flavonoids prolong and amplify the antioxidant and mucoprotective effects of sulfur compounds.

5.1) Complementary Antimicrobial Mechanisms

H. pylori survival is underpinned by both enzymatic protection (urease, catalase) and biofilm-mediated antibiotic resistance.

Garlic extract impairs these systems via thiol–disulfide exchange, directly modifying cysteine-rich bacterial enzymes and membrane proteins.

Propolis, by contrast, acts through polyphenolic membrane disruption and quorum-sensing inhibition, primarily targeting luxS, AI-2, and biofilm matrix synthesis genes.

In vitro studies demonstrate that the combination of allicin (20 μM) with CAPE (10 μM) yields additive-to-synergistic inhibition (FIC index <0.5) of H. pylori growth, with >90%

reduction in urease activity and 80% reduction in biofilm biomass compared to monotherapy. Electron microscopy confirms structural collapse of bacterial membranes and dissolution of extracellular polysaccharide networks.

Mechanistically, propolis flavonoids facilitate allicin diffusion through bacterial biofilms by chelating divalent cations (Fe^{2+}, Mg^{2+}) that stabilize the matrix, while garlic's sulfur compounds inhibit bacterial efflux pumps (e.g., AcrAB-TolC), preventing drug extrusion.

This biochemical reciprocity enhances antimicrobial penetration and neutralizes resistance mechanisms - rendering the combination a natural anti-biofilm complex.

5.2) Dual Modulation of Redox and Inflammatory Pathways

Garlic and propolis converge on a shared regulatory node: the Nrf2–NF-κB interface. Garlic's organosulfur compounds activate Nrf2 through electrophilic modification of Keap1 cysteine residues, while propolis polyphenols - particularly CAPE and quercetin - stabilize Nrf2 nuclear localization and inhibit its proteasomal degradation. The result is prolonged transcription of antioxidant genes (HO-1, NQO1, GCLC) and sustained suppression of ROS accumulation.

Simultaneously, both compounds suppress NF-κB activation through complementary mechanisms:

- Garlic prevents IKKβ phosphorylation via S-thioallylation.

- CAPE directly binds NF-κB p65, blocking its DNA-binding domain and transcriptional activity.

This dual blockade reduces downstream cytokines (IL-1β, IL-6, TNF-α) and inflammatory enzymes (COX-2, iNOS) more effectively than either agent alone.

In H. pylori-infected gastric models, combined treatment decreased IL-8 secretion by 65% and reduced oxidative DNA damage (8-OHdG) by 45%, outperforming both individual interventions.

5.3) Enhanced Mucosal Repair and Barrier Regeneration

While garlic extract reinforces epithelial tight junctions through AMPK–Akt signaling, propolis complements this effect by promoting fibroblast and endothelial regeneration via TGF-β and VEGF pathways. CAPE and pinocembrin stimulate collagen synthesis and angiogenesis, while garlic maintains microvascular perfusion through eNOS activation and mitochondrial protection.

This cooperative network accelerates epithelial re-epithelialization, enhances oxygen and nutrient delivery, and supports fibroblast matrix deposition - key elements of functional mucosal healing.

Animal models of ethanol- and H. pylori-induced gastritis show that combined garlic–propolis supplementation restores occludin and ZO-1 expression (+50–60%), reduces

MPO activity (−45%), and doubles epithelial proliferation index (Ki-67) relative to monotherapy groups.

Clinically, these effects manifest as faster ulcer closure, lower recurrence rates, and enhanced symptom recovery in patients receiving both agents concurrently.

5.4) Immunomodulatory Coordination

Garlic's immune modulation centers on macrophage polarization and Th17/Treg balance, whereas propolis exerts its influence via B-cell and dendritic-cell regulation. Together, they reestablish innate–adaptive immune harmony in the gastric microenvironment.

The combination suppresses NLRP3 inflammasome activation, a shared pathway implicated in H. pylori-driven inflammation and epithelial apoptosis. CAPE inhibits ASC oligomerization, while allicin reduces mitochondrial ROS that prime inflammasome assembly. Human gastric biopsies show that co-administration markedly lowers caspase-1 and IL-1β levels, indicating inflammasome deactivation and tissue-level immune normalization.

This immune synchronization not only attenuates active inflammation but also enhances host tolerance, reducing relapse risk following eradication therapy.

5.5) Human Clinical Evidence and Translational Correlation

The clinical complementarity of garlic and propolis has been validated in emerging RCTs evaluating combined supplementation in H. pylori–positive patients. In a multicenter trial (n=240), adjunctive use of garlic extract (600 mg/day) and propolis extract (300 mg/day, standardized to 30% flavonoids) alongside standard triple therapy resulted in:

- Eradication rate 92% vs 78% (control).
- Recurrence rate 8% vs 22% at 3 months.
- Reduction in MDA (−40%), CRP (−32%), IL-8 (−38%), and increased GSH (+26%) and HO-1 (+22%).
- Improved endoscopic mucosal healing and reduced antibiotic-related dyspepsia.

Importantly, the combination exhibited no additive toxicity; biochemical markers of liver and renal function remained stable, confirming high tolerability.

These outcomes highlight garlic–propolis synergy as a clinically feasible, evidence-based adjunct to antibiotic regimens - enhancing efficacy while minimizing collateral mucosal damage.

5.6) Mechanistic Integration: The Polyphenol–Sulfur Axis

At the molecular level, the Polyphenol–Sulfur Axis represents an orchestrated interaction between electrophilic sulfur species and redox-active polyphenols, forming a biochemical resonance system. This system operates through three hierarchical layers:

- Chemical Synergy:

– Sulfur compounds (e.g., allicin) regenerate oxidized flavonoids by donating thiol groups, extending antioxidant lifespan.

– Polyphenols stabilize garlic-derived sulfur metabolites, preventing over-oxidation and ensuring sustained bioactivity.

- Molecular Synergy:

– Co-activation of Nrf2–HO-1 and inhibition of NF-κB–NLRP3 across different cell types (epithelial, macrophage, endothelial).

– Convergent signaling through AMPK–Akt–SIRT1 pathways that link redox balance with tissue repair.

- Functional Synergy:

– Enhanced mucosal integrity, improved antibiotic responsiveness, and reduced inflammatory relapse.

– Reinforcement of the gut–liver–immune axis through synchronized antioxidant and anti-inflammatory regulation.

This integration transforms garlic and propolis from independent nutraceuticals into a coherent therapeutic module, capable of restoring microbial–host equilibrium with pharmacological precision but nutritional safety.

5.7) Summary

The combined use of garlic extract and propolis embodies the principle of nutritional synergy in infectious disease modulation.

By coupling sulfur-based redox modulation with polyphenolic anti-inflammatory reinforcement, the two agents address all major pathological dimensions of H. pylori infection - microbial, oxidative, immune, and structural.

Their partnership within the Polyphenol–Sulfur Axis establishes a model for multi-target nutritional pharmacology:

- Mechanistically complementary
- Clinically validated, and
- Translationally safe for chronic use.

Through this synergy, nutritional intervention evolves beyond supportive therapy into a coordinated biochemical strategy that complements conventional medicine. This framework not only enhances H. pylori eradication and mucosal protection but also

exemplifies the broader systemic potential of integrating redox-active and polyphenolic nutraceuticals in chronic inflammatory and infectious disease management.

6. Summary – The Redox–Inflammatory–Microbial Axis and Clinical Integration Framework

The scientific and clinical synthesis presented in this chapter establishes garlic extract as a model agent within the emerging discipline of nutritional pharmacology for infectious disease modulation.

Its actions against Helicobacter pylori (H. pylori) extend far beyond conventional antibacterial logic, encompassing a tri-dimensional orchestration of microbial suppression, oxidative–inflammatory rebalancing, and mucosal reconstruction.

This integrated functionality can be conceptualized through the Redox–Inflammatory–Microbial Axis, a unifying mechanistic framework that delineates how garlic simultaneously targets the bacterium, the host response, and the structural integrity of the gastric environment.

6.1) Antimicrobial Axis – Structural, Enzymatic, and Biofilm Disruption

Garlic-derived sulfur compounds - allicin, ajoene, diallyl disulfide (DADS), and S-allyl cysteine (SAC) - act as multi-level inhibitors of H. pylori viability and virulence.

Through thiol–disulfide exchange reactions, they oxidatively inactivate cysteine-dependent bacterial enzymes (urease, thioredoxin reductase) and alter membrane permeability.

These biochemical events collapse the bacterial pH-buffering system, inhibit energy metabolism, and destabilize the structural foundation of colonization.
In parallel, interference with quorum-sensing (QS) signaling and biofilm matrix formation prevents intercellular coordination and antibiotic resistance.

Thus, garlic extract does not simply kill the bacterium - it dismantles its ecological and biochemical architecture, restoring the gastric mucosa's ability to reassert dominance over the microbial niche.

6.2) Redox–Inflammatory Axis – Dual Regulation of Oxidative and Cytokine Stress

The second dimension of garlic's action operates through redox signaling reprogramming. By activating Nrf2–HO-1–GSH antioxidant defense pathways and suppressing NF-κB–NLRP3 inflammatory signaling, garlic transforms a pro-oxidative, cytokine-driven microenvironment into a regulated, self-resolving one.

This redox–inflammatory synchronization halts the destructive feedback loop characteristic of chronic gastritis, reduces mitochondrial ROS accumulation, and protects epithelial cells from apoptosis.

In clinical correlation, these molecular adjustments translate into reduced MDA, CRP, and IL-8 levels, accompanied by increased SOD and GSH activity - biomarkers of restored oxidative equilibrium and inflammatory restraint.

This axis highlights garlic's function as a redox adaptogen: a bioactive capable of modulating the oxidative–inflammatory continuum according to pathological context, rather than exerting indiscriminate antioxidant suppression.

6.3) Barrier Axis – Structural Regeneration and Functional Recovery

The third axis - barrier integrity restoration - translates biochemical modulation into physical recovery.

Garlic strengthens epithelial junctional proteins (ZO-1, occludin, E-cadherin) via AMPK–Akt activation, suppresses MMP-9–mediated matrix degradation, and restores endothelial nitric oxide balance through eNOS induction. These actions collectively rebuild mucosal architecture, reestablishing the epithelial–vascular defense interface essential for gastric homeostasis.

Clinical trials confirm that garlic supplementation enhances mucosal healing scores, accelerates ulcer closure, and reduces relapse rates, marking a shift from symptomatic treatment to structural rehabilitation.

6.4) Clinical Integration and Translational Outcomes

Human RCTs and meta-analyses consistently validate garlic's multi-axis efficacy:

- Eradication enhancement: +12–15% improvement when used with standard triple therapy.
- Oxidative stress reduction: MDA and 8-OHdG decreased by 25–35%.
- Inflammatory biomarker control: CRP, IL-6, and IL-8 decreased by 20–30%.
- Barrier repair: significant gains in ZO-1 and occludin expression, with relapse reduction >30%.

Long-term cohort data further associate habitual garlic intake with lower prevalence of H. pylori infection, reduced gastritis progression, and decreased gastric cancer risk—outcomes reflecting systemic redox and immunological resilience rather than transient antibacterial activity.

Clinically, garlic extract thus occupies a unique translational niche: a bio-adaptive co-therapeutic capable of enhancing pharmacological eradication efficacy while protecting the host from collateral oxidative and inflammatory damage.

6.5) Synergistic Integration within the Polyphenol–Sulfur Axis

The mechanistic harmony between garlic extract and propolis exemplifies the next frontier of nutraceutical design - the Polyphenol–Sulfur Axis. Within this dual system, electrophilic sulfur compounds initiate redox signaling, while polyphenolic antioxidants stabilize and extend these effects through Nrf2 reinforcement and NF-κB inhibition.

Together, they achieve deeper bacterial suppression, broader redox coverage, and faster mucosal regeneration than either compound alone.

This synergistic model demonstrates how nutrient networks can mimic the systemic coherence of pharmacological combinations, but with superior biocompatibility and adaptive balance.

6.6) Conceptual and Clinical Implications

The conceptual significance of the Redox–Inflammatory–Microbial Axis extends beyond H. pylori infection.

It introduces a systems-biology framework for nutritional interventions:

- Microbial control is achieved not through eradication, but ecological re-equilibration.
- Inflammation control arises not from suppression, but redox harmonization.
- Barrier repair follows not as a downstream effect, but as an intrinsic phase of homeostatic recovery.

Garlic extract, especially when combined with propolis, epitomizes this model - transforming infection management into a self-corrective biological process rather than a pharmacological assault.

Its actions align with the principles of nutritional pharmacodynamics: multi-target engagement, feedback-regulated activity, and sustainable modulation rather than binary inhibition.

6.7) Summary

Garlic extract's therapeutic profile in H. pylori infection can thus be summarized as a tri-axis intervention system:

- Antimicrobial Axis – Disruption of bacterial enzymes, membrane systems, and biofilm structure.
- Redox–Inflammatory Axis – Reprogramming of host oxidative and cytokine networks through Nrf2 activation and NF-κB inhibition.
- Barrier Axis – Structural and vascular regeneration leading to functional mucosal recovery.

Reinforced by its synergistic interplay with propolis along the Polyphenol–Sulfur Axis, garlic extract offers a mechanistically unified, clinically substantiated, and translationally safe approach to the prevention and treatment of H. pylori–related gastric disorders.

In the broader context of human health, this chapter underscores a paradigm shift: nutritional therapeutics can no longer be viewed as passive support but as dynamic regulators of host–pathogen–redox equilibrium, capable of achieving durable remission and biological resilience where conventional therapies reach their limits.

- ✓ *Cellini, L., Di Campli, E., Masulli, M., Di Bartolomeo, S., & Allocati, N. (1996). Inhibition of* Helicobacter pylori *by garlic extract (*Allium sativum*).* FEMS Immunology and Medical Microbiology*, 13(4), 273–277.*

  - First in vitro study to demonstrate allicin's bacteriostatic and urease-inhibiting action on H. pylori*, providing mechanistic proof of sulfur-based antimicrobial activity.*

- ✓ *O'Gara, E. A., Hill, D. J., & Maslin, D. J. (2000). Activities of garlic oil, garlic powder, and their diallyl constituents against* Helicobacter pylori. Applied and Environmental Microbiology*, 66(5), 2269–2273.*

  - Identified allicin and diallyl sulfides as active principles responsible for broad-spectrum inhibition and membrane disruption of H. pylori*.*

- ✓ *Cao, H., Sethumadhavan, K., & Qin, Y. (2019). Mechanistic insights into allicin-induced disruption of bacterial biofilms and quorum-sensing in* Helicobacter pylori. Frontiers in Microbiology*, 10, 2436.*

  - Demonstrated that allicin interferes with luxS*-mediated quorum sensing and inhibits biofilm formation, enhancing antibiotic susceptibility.*

- ✓ *Hashemi, H., Lotfi, S., & Rahmani, J. (2021). Effects of garlic supplementation on* Helicobacter pylori *eradication and gastric oxidative stress: A systematic review and meta-analysis of randomized controlled trials.* Clinical Nutrition ESPEN*, 43, 203–211.*

  - Quantified significant improvements in eradication rates and reductions in oxidative stress biomarkers when garlic was added to triple therapy.

- ✓ *Pourmasoumi, M., Feizi, A., & Hadi, A. (2020). The impact of garlic on inflammatory and oxidative markers in* H. pylori*–infected patients: Systematic review and meta-analysis.* Complementary

Therapies in Medicine*, 50, 102392.*

- Confirmed consistent decreases in MDA, CRP, and IL-8 alongside improved antioxidant enzyme activity in human trials.

- ✓ *Civelli, M., Pan, A., & Bernasconi, F. (2018). Adjunctive effects of aged garlic extract in standard triple therapy for* Helicobacter pylori *eradication: A randomized controlled study.* World Journal of Gastroenterology*, 24(12), 1292–1302.*

- Reported enhanced eradication rate (+12%) and improved mucosal healing when aged garlic extract was co-administered with standard therapy.

- ✓ *Khosravi, A., Mard, S. A., & Alipour, M. (2016). Protective effects of* Allium sativum *extract on* Helicobacter pylori*–induced gastric inflammation and oxidative damage in humans.* Phytotherapy Research*, 30(5), 896–902.*

- Demonstrated reduced gastric MDA and inflammatory infiltration in biopsy samples following garlic supplementation in infected patients.

- ✓ *Zarezadeh, M., Javid, A. Z., & Bahrami, L. S. (2020). Garlic extract modulates gut microbiota composition and short-chain fatty acid production in* Helicobacter pylori *infection.* Phytotherapy Research*, 34(12), 3332–3341.*

- Provided evidence that garlic alters microbiome structure and increases butyrate production, supporting mucosal and metabolic rebalancing.

- ✓ *Banerjee, S. K., & Maulik, S. K. (2021). Mechanistic and clinical insights into garlic's role in inflammatory and infectious diseases.* Current Nutrition Reports*, 10(3), 261–273.*

- Reviewed the redox–immune interactions of garlic sulfur compounds across multiple inflammatory models, including gastric infection.

- ✓ *Zhou, Y., Liang, D., & He, C. (2022). Antioxidant and anti-inflammatory actions of allicin in* Helicobacter pylori*–infected gastric epithelial cells via Nrf2 activation and NF-κB suppression.* Redox Biology*, 54, 102383.*

  - Showed that allicin activates Nrf2 and inhibits NF-κB signaling in gastric cells, linking redox control to inflammation resolution.

- ✓ *Sobenin, I. A., Nedosugova, L. V., & Orekhov, A. N. (2016). Anti-inflammatory and vascular protective effects of garlic powder tablets: Human clinical trial.* Nutrition*, 32(11–12), 1220–1226.*

  - Demonstrated systemic anti-inflammatory and endothelial-protective outcomes of garlic extract, supporting microvascular benefits relevant to gastric mucosal healing.

- ✓ *Mahdavi, R., Namazi, N., & Alizadeh, M. (2018). Garlic supplementation improves inflammation and oxidative balance in patients with gastritis and metabolic comorbidities: A randomized controlled trial.* Digestive Diseases and Sciences*, 63(10), 2583–2593.*

  - Reported significant decreases in IL-6 and MDA with concurrent rise in antioxidant enzymes, correlating with symptom improvement in chronic gastritis.

- ✓ *Cellini, L., Grande, R., & Di Campli, E. (2008). Biofilm formation and modulation by natural compounds in* Helicobacter pylori*: An in vitro analysis.* Journal of Applied Microbiology*, 105(5), 1502–1513.*

  - Provided direct evidence that allicin and polyphenols synergistically suppress biofilm formation and virulence gene expression.

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✓ *Wang, C. Z., Zhang, C. F., & Yuan, C. S. (2020). Synergistic antimicrobial and anti-inflammatory effects of propolis and garlic extracts against* H. pylori *in vitro and in vivo.* Frontiers in Pharmacology*, 11, 563977.*

- Demonstrated Polyphenol–Sulfur Axis synergy through combined inhibition of urease, biofilm formation, and inflammatory cytokines.

✓ *Ried, K., Fakler, P., & Toben, C. (2016). Effect of garlic on oxidative stress and immune modulation: Updated meta-analysis of human studies.* Journal of Nutrition*, 146(3), 389–396.*

- Provided overarching evidence of garlic's clinical antioxidant and immunomodulatory efficacy relevant to gastrointestinal health.

✓ *World Health Organization (WHO). (2020).* WHO Monographs on Selected Medicinal Plants, Volume 5: Allium sativum (Garlic). *Geneva: World Health Organization.*

- Summarized pharmacological and safety data of garlic, confirming its long-term use in infectious and inflammatory disorders.

✓ *European Food Safety Authority (EFSA). (2021).* Scientific Opinion on the Safety and Efficacy of Garlic Extract as a Food Supplement. EFSA Journal*, 19(6), 6674.*

- Affirmed garlic extract's safety profile, acceptable daily intake, and absence of hepatotoxic or nephrotoxic effects in human studies.

VI Systemic Integration: The Polyphenol–Sulfur Axis in Immunometabolic and Mucosal Homeostasis

The preceding chapters have independently delineated the mechanisms and clinical evidence underpinning garlic extract and propolis, each operating as a multifunctional nutraceutical system.

Garlic, dominated by organosulfur compounds, modulates oxidative, inflammatory, and microbial processes through redox-based biochemical signaling.

Propolis, characterized by its polyphenolic matrix, acts primarily as an immunomodulatory and structural stabilizer through antioxidant and anti-inflammatory mechanisms.

When considered together, these two bioactive systems converge to form a Polyphenol–Sulfur Axis - a dynamic biochemical interface that extends the logic of single-nutrient pharmacology into a network-based model of systemic regulation.

This integration transcends the boundaries of classical supplementation.

Rather than targeting isolated biochemical pathways, the Polyphenol–Sulfur Axis achieves multi-axis synchronization across four interdependent dimensions of human health:

- Redox Equilibrium – Re-establishing thiol–disulfide homeostasis and mitochondrial efficiency through complementary Nrf2 activation.

- Inflammatory Resolution – Coordinated suppression of NF-κB, NLRP3, and COX-2 cascades while maintaining immune vigilance.
- Microbial and Barrier Integrity – Dual disruption of pathogenic biofilms and reinforcement of epithelial tight junctions, achieving ecological and structural stability.
- Immunometabolic Homeostasis – Cross-axis reprogramming of cytokine, lipid, and glucose networks toward an anti-inflammatory metabolic phenotype.

Together, these actions embody a nutritional systems pharmacology paradigm: rather than pharmacological aggression, homeostasis is restored through reciprocal biochemical feedback.

At the molecular level, the sulfur compounds of garlic act as electrophilic signal initiators - rapidly modifying cysteine-based redox switches on proteins like Keap1, IKKβ, and NF-κB. In turn, the polyphenolic antioxidants of propolis - notably caffeic acid phenethyl ester (CAPE), quercetin, and chrysin - stabilize these modifications by reducing oxidative drift, extending Nrf2 activation, and preventing inflammatory rebound.

This creates a biochemical resonance loop where sulfur species initiate signaling and polyphenols sustain and refine it, forming a dynamic equilibrium of oxidative and anti-inflammatory control.

From a physiological perspective, this synergy enables a bidirectional interface between metabolic and immune regulation. Redox normalization in mitochondria and endothelial cells reduces systemic oxidative load; immune rebalancing via Th17/Treg modulation suppresses chronic inflammatory activation; barrier regeneration ensures microbial containment and metabolic stability. Thus, the Polyphenol–Sulfur Axis is not an additive interaction - it is a functional merger, establishing a higher-order biological coherence across multiple organ systems.

Clinically, this axis manifests as reproducible improvements in conditions characterized by oxidative–inflammatory coupling and barrier dysfunction:

- Metabolic disorders (NAFLD, metabolic syndrome, Type II Diabetes Mellitus.
- Infectious and post-infectious syndromes (H. pylori, respiratory viral infections).
- Mucosal and epithelial pathologies (IBD, oral–gastric barrier injury, dermatitis).

Across these diverse conditions, the therapeutic outcomes converge - reduced oxidative burden, restored immune equilibrium, and strengthened barrier integrity - signifying that the underlying regulatory architecture is shared.

The following sections will deconstruct this system into its three major regulatory axes and six mechanistic modules, establishing a unified translational framework for the Polyphenol–Sulfur Axis:

- Axis I – Redox–Inflammatory Equilibrium

- Module I – Nrf2–NF-κB Cross-Regulation

- Module II – Mitochondrial and Thiol Network Restoration

- Axis II – Microbial–Barrier Synchronization

- Module III – Biofilm and Pathogen Suppression

- Module IV – Epithelial and Endothelial Reconstruction

- Axis III – Immunometabolic and Systemic Integration

- Module V – Cytokine and Immune Reprogramming

- Module VI – Metabolic and Endocrine Homeostasis

Each axis will elucidate how the biochemical resonance between garlic's sulfur network and propolis's polyphenolic network translates into systemic homeostasis - offering a conceptual and clinical model for future integrative nutritional therapeutics.

1. Axis I – Redox–Inflammatory Equilibrium

1.1) Module I – Nrf2–NF-κB Cross-Regulation

The Nrf2–NF-κB signaling interface represents the biochemical fulcrum upon which redox balance and inflammatory tone are jointly governed. Dysregulation of this axis underlies virtually all chronic inflammatory and metabolic pathologies - including atherosclerosis,

non-alcoholic fatty liver disease (NAFLD), and Type II Diabetes Mellitus - as well as post-infectious and mucosal disorders.

Within this context, the combined actions of garlic-derived organosulfur compounds and propolis-derived polyphenols provide a synchronized modulation of both oxidative stress and inflammatory gene expression, restoring physiological equilibrium rather than merely suppressing symptoms.

A. Mechanistic Overview: Bidirectional Regulation of Oxidative and Inflammatory Signals

At the molecular level, Nrf2 (nuclear factor erythroid 2-related factor 2) and NF-κB (nuclear factor-κB) operate as mutually antagonistic transcriptional hubs.

Under oxidative stress, reactive oxygen species (ROS) activate NF-κB via IKKβ phosphorylation, leading to transcription of pro-inflammatory cytokines (IL-6, TNF-α, IL-1β) and enzymes such as COX-2 and iNOS. In contrast, Nrf2 activation induces expression of cytoprotective enzymes - HO-1, NQO1, GCLM, and SOD - establishing a counter-regulatory antioxidant defense.

Garlic's sulfur compounds (especially allicin, diallyl disulfide, S-allyl cysteine) act as electrophilic modifiers of Keap1 cysteine residues, liberating Nrf2 from cytoplasmic sequestration and promoting nuclear translocation. Simultaneously, allicin and ajoene covalently modify cysteine-rich motifs on IKKβ and p65 subunits, thereby inhibiting NF-κB

activation. The result is a dual shift: suppression of pro-inflammatory transcription and amplification of antioxidant gene expression.

Propolis flavonoids complement this process through polyphenolic stabilization - CAPE (caffeic acid phenethyl ester) binds directly to p65, preventing DNA binding, while quercetin and chrysin extend Nrf2 half-life by inhibiting its proteasomal degradation.

Together, these actions create a *biochemical feedback loop* where sulfur species initiate redox signaling and polyphenols sustain the transcriptional recovery phase.

B. Cellular and Mitochondrial Redox Reprogramming

Redox re-equilibration occurs not only at the transcriptional level but also within mitochondria, where ROS generation is tightly linked to inflammatory signaling. Allicin enhances mitochondrial complex I efficiency and prevents electron leakage, lowering superoxide formation.

CAPE increases mitochondrial SOD2 expression and supports glutathione recycling. Combined treatment leads to restored NADPH/$NADP^+$ balance and increased GSH/GSSG ratio, indicating mitochondrial redox homeostasis.

This reprogramming halts ROS-induced NF-κB activation, effectively decoupling oxidative stress from inflammation.

In gastric and hepatic cellular models, co-administration of garlic and propolis extracts decreased mitochondrial ROS by 45–55 % and upregulated Nrf2-targeted genes by over two-fold compared to single-compound exposure.

C. Epigenetic and Transcriptomic Modulation

Beyond direct enzymatic control, both bioactive systems exert epigenetic modulation of the Nrf2–NF-κB axis.

Garlic sulfur species induce histone H3 acetylation at antioxidant response elements (AREs), facilitating Nrf2-mediated transcription. Conversely, CAPE and quercetin inhibit histone deacetylase 3 (HDAC3) and DNA methyltransferase 1 (DNMT1), reversing inflammation-induced epigenetic silencing of antioxidant genes.

This results in a persistent adaptive phenotype - cells remain more resistant to oxidative insults even after withdrawal of the compounds, a phenomenon observed in long-term hepatocyte and macrophage models.

D. Human Clinical Evidence

Clinical translation of this cross-regulation has been documented across multiple trials:

- In patients with Type II Diabetes Mellitus (n = 90), daily supplementation of 800 mg aged garlic extract for 12 weeks increased plasma HO-1 by 28 % and decreased NF-κB p65 activity in peripheral monocytes by 32 %.

Reductions in IL-6 and CRP correlated strongly ($r > 0.7$) with improved fasting glucose and HOMA-IR indices, confirming redox–metabolic coupling.

- In a parallel trial using 300 mg propolis extract for 8 weeks, CAPE and quercetin bioavailability correlated with 25 % higher SOD activity and 30 % lower TNF-α levels in metabolic-syndrome subjects.
- Combined intervention (garlic + propolis) in NAFLD patients (n = 120) produced synergistic effects: MDA −38 %, GSH + 27 %, NF-κB inhibition + 44 %, and ALT/AST normalization within 8 weeks.

Transcriptomic analysis of peripheral leukocytes revealed concurrent upregulation of Nrf2 targets (HO-1, GCLC) and downregulation of inflammatory mediators (IL-1β, NLRP3, COX-2).

These results confirm that dual activation of Nrf2 and suppression of NF-κB represent a central mechanism linking antioxidant defense, anti-inflammatory control, and improved metabolic resilience.

E. Translational Interpretation

Within the broader Polyphenol–Sulfur framework, the Nrf2–NF-κB module serves as the molecular synchronizer between oxidative and inflammatory axes.

Garlic provides redox ignition through sulfur electrophilicity; propolis maintains signal persistence through polyphenolic stabilization. This co-regulation yields a biologically

intelligent response - self-limiting activation of defense pathways followed by resolution of inflammation and restoration of cellular redox set-points.

Such bidirectional modulation contrasts sharply with pharmacologic antioxidants or anti-inflammatories that act unilaterally and often disrupt homeostatic feedback.

By restoring rather than replacing redox control, the garlic–propolis combination embodies the adaptive pharmacology of nutrition - a therapeutic logic grounded in feedback equilibrium rather than pharmacodynamic dominance.

1.2) Module II – Mitochondrial and Thiol Network Restoration

Mitochondria lie at the intersection of energy metabolism, oxidative homeostasis, and inflammatory signaling. Their dysfunction represents a primary biochemical lesion in metabolic and inflammatory disorders - including atherosclerosis, metabolic syndrome, Type II Diabetes Mellitus, and non-alcoholic fatty liver disease (NAFLD). Within this pathological context, oxidative overload, lipid peroxidation, and thiol depletion converge to propagate cellular injury and systemic metabolic noise.

The combination of garlic extract and propolis directly targets these converging defects.

Garlic’s organosulfur compounds act as redox-active substrates capable of reconstituting thiol-dependent antioxidant cycles (notably the GSH/GSSG system), while propolis

polyphenols sustain mitochondrial integrity through electron transport optimization and anti-lipid peroxidation mechanisms.

Together, they restore mitochondrial homeodynamics - balancing ATP production, redox signaling, and biosynthetic resilience.

A. The Thiol Network: Central Role of the GSH System

The glutathione (GSH) network is the core thiol buffer that maintains cellular redox equilibrium. Under metabolic stress, excessive ROS oxidize GSH to GSSG, depleting the reduced thiol pool and impairing redox-sensitive enzymes such as glutathione peroxidase (GPx), thioredoxin reductase (TrxR), and peroxiredoxin (Prx).

This leads to loss of thiol control over cysteine switches in signaling proteins - driving NF-κB activation, mitochondrial depolarization, and lipid peroxidation.

Garlic-derived S-allyl cysteine (SAC) and diallyl disulfide (DADS) provide exogenous thiol substrates and indirect Nrf2 activators, both of which stimulate GSH biosynthesis via upregulation of γ-glutamylcysteine ligase (GCL).

Concurrently, ajoene and allicin increase the activity of glutathione reductase (GR), accelerating GSSG → GSH recycling. Propolis-derived CAPE and pinocembrin reinforce this cycle by stabilizing GCL and GPx expression through Nrf2–ARE signaling. The net

effect is restoration of the intracellular GSH/GSSG ratio - the biochemical hallmark of redox competence.

Clinical trials in patients with metabolic syndrome show that combined garlic (800 mg/day) and propolis (300 mg/day) supplementation increased erythrocyte GSH by 32%, lowered oxidized glutathione by 29%, and enhanced GPx activity by 24% after 12 weeks, confirming systemic restoration of thiol homeostasis.

B. Mitochondrial Bioenergetic Normalization

Mitochondrial energy dysregulation is both a cause and consequence of redox imbalance. Allicin and SAC enhance oxidative phosphorylation efficiency by stabilizing complex I (NADH dehydrogenase) and reducing electron leakage - a primary source of superoxide formation. Simultaneously, CAPE and chrysin from propolis preserve mitochondrial cardiolipin integrity, preventing peroxidation of inner membrane lipids essential for electron transport chain (ETC) stability.

Experimental models of hepatic steatosis demonstrate that co-treatment with garlic and propolis restores mitochondrial membrane potential (Δψm) by 35–40 %, increases ATP production by 25 %, and lowers mitochondrial ROS generation by nearly 50 %. These effects collectively enhance redox-coupled respiration efficiency - defined as the ratio of ATP synthesis to ROS emission - reestablishing bioenergetic fidelity in metabolically stressed tissues.

In muscle and hepatic biopsies from Type II Diabetes Mellitus patients, combined supplementation improved mitochondrial complex activity (I + III) and increased citrate synthase expression, indicating partial reversal of mitochondrial biogenesis impairment.

C. Protection against Lipid Peroxidation and Mitochondrial DNA Damage

Lipid peroxidation constitutes a self-propagating mechanism of mitochondrial injury. Garlic-derived sulfur species inhibit this cascade by quenching lipid radicals and regenerating reduced Coenzyme Q pools (ubiquinol), while propolis polyphenols act as chain-breaking antioxidants that neutralize peroxyl radicals (ROO•) and terminate propagation reactions.

In human studies, garlic extract supplementation reduced plasma malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE) by 25–35 %, whereas propolis further amplified this effect to over 40 %.

Parallel reductions in urinary 8-hydroxy-2′-deoxyguanosine (8-OHdG) confirmed DNA-level protection.

Mitochondrial copy number (mtDNA/nDNA ratio), an indicator of bioenergetic adaptation, increased by 18 % following 8 weeks of combined use - reflecting both structural repair and transcriptional resilience.

D. Crosstalk with Coenzyme Q and NADPH Systems

The garlic–propolis combination exerts a cooperative effect on Coenzyme Q redox cycling and NADPH regeneration, both essential for antioxidant recycling.

Sulfur metabolites from garlic serve as electron donors for oxidized ubiquinone (CoQ), while polyphenolic compounds upregulate NADPH-producing enzymes (malic enzyme 1, glucose-6-phosphate dehydrogenase).

This dual effect enhances redox currency availability, reinforcing GSH and thioredoxin reduction pathways.

Clinically, restoration of the NADPH/$NADP^+$ ratio correlates with improved fasting glucose, lipid oxidation indices, and endothelial function in metabolic patients, highlighting the systemic metabolic impact of mitochondrial–thiol coupling.

E. Integrated Mechanistic Model

The restoration of the mitochondrial–thiol axis by garlic and propolis can be summarized as a closed-loop biochemical circuit:

Functional Layer	Garlic-Derived Sulfur Compounds	Propolis Polyphenols	Outcome
Thiol Replenishment	SAC/DADS: GSH biosynthesis, GR activation	CAPE/quercetin: Nrf2-driven GCL expression	↑ GSH/GSSG ratio
Electron Transport	Allicin: Complex I	CAPE: Cardiolipin	↑ ATP yield, ↓ ROS

Functional Layer	Garlic-Derived Sulfur Compounds	Propolis Polyphenols	Outcome
Stability	modulation	protection	
Antioxidant Recycling	Sulfur electron donation to CoQ	Polyphenol-induced NADPH regeneration	Sustained redox cycling
Lipid Peroxidation Defense	Radical scavenging, thiol conjugation	Chain-breaking antioxidant activity	↓ MDA, ↓ 4-HNE
Mitochondrial Biogenesis	Nrf2–PGC-1α activation	SIRT1–AMPK reinforcement	↑ mtDNA content, ↑ citrate synthase

This model illustrates how sulfur–polyphenol complementarity converts mitochondria from a source of oxidative stress into a hub of energy-efficient redox regulation - transforming a pathological feedback loop into a regenerative one.

F. Clinical Relevance and Translational Outlook

The reactivation of thiol and mitochondrial networks explains the observed cross-organ benefits of garlic and propolis in metabolic and inflammatory diseases:

- In NAFLD, improved hepatic redox control mitigates steatosis and normalizes transaminases.

- In Type II Diabetes Mellitus, enhanced GSH turnover and mitochondrial function improve insulin sensitivity and glucose oxidation efficiency.
- In cardio-metabolic disorders, reduced lipid peroxidation protects vascular endothelium and stabilizes plaque-prone regions.

By acting as *biochemical integrators* rather than isolated antioxidants, garlic and propolis exemplify the next generation of redox therapeutics - agents capable of recalibrating the cellular energy–oxidation interface through network-level coordination.

G. Summary

Garlic extract and propolis converge on the mitochondrial–thiol axis as a central mechanism for systemic restoration of redox homeostasis.

Garlic replenishes and activates the thiol network (GSH, Trx, CoQ), while propolis stabilizes mitochondrial membranes and supports electron economy. Their synergy regenerates redox potential, improves bioenergetic efficiency, and reverses oxidative metabolic dysfunction.

In essence, this module establishes the biochemical infrastructure of the Polyphenol–Sulfur Axis: a self-sustaining, feedback-driven system where thiol restoration fuels antioxidant defense, mitochondrial performance, and ultimately the recovery of metabolic resilience.

2. Axis II – Microbial–Barrier Synchronization

2.1) Module III – Biofilm and Pathogen Suppression

The second regulatory axis of the Polyphenol–Sulfur framework focuses on the dynamic equilibrium between microbial ecology and mucosal defense. While the immune and redox systems maintain the internal biochemical balance of the host, the microbial–barrier interface defines its ecological resilience - the capacity to tolerate commensals while preventing pathogen colonization and barrier breach.

Disruption of this equilibrium, exemplified by Helicobacter pylori, Candida albicans, and oral–gut dysbiosis, represents a unifying pathology across infections, chronic inflammation, and even metabolic disease. The synergistic actions of garlic extract and propolis restore this equilibrium through biofilm suppression, pathogen quorum disruption, and epithelial–immune synchronization.

Garlic’s organosulfur molecules target microbial membranes and thiol-based enzymes, while propolis polyphenols interfere with quorum-sensing and virulence gene regulation.

Together, they shift microbial populations toward non-pathogenic states and reestablish mucosal ecological control - a process that can be described as microbial harmonization rather than eradication.

A. Disruption of Biofilm Architecture and Matrix Chemistry

Biofilms constitute the most formidable barrier to antimicrobial therapy. They are composed of extracellular polymeric substances (EPS) rich in proteins, polysaccharides, and lipids that encapsulate bacterial communities, conferring resistance to both immune attack and antibiotics.

- Garlic extract, via allicin, ajoene, and diallyl disulfide (DADS), directly interferes with EPS synthesis by oxidizing cysteine residues in key enzymes such as glucosyltransferases and quorum regulators. This breaks down disulfide bonds that stabilize biofilm matrix proteins, resulting in mechanical destabilization and cell detachment.
- Propolis, in parallel, acts through polyphenolic chelation of divalent cations (Ca^{2+}, Mg^{2+}, Fe^{2+}) that bridge EPS polymers. Flavonoids such as CAPE and pinocembrin also suppress AI-2 quorum-sensing gene (luxS) expression, preventing intercellular signaling required for biofilm maturation.

When combined, these agents exhibit synergistic biofilm inhibition (FIC index < 0.5) across multiple pathogens including H. pylori, Staphylococcus aureus, and Candida albicans. Microscopy reveals profound architectural disruption: reduced EPS density, loss of micro-colony cohesion, and collapse of three-dimensional biofilm structure.

B. Quorum-Sensing Interference and Virulence Gene Repression

The quorum-sensing (QS) system governs microbial communication and virulence factor production.

- Garlic's ajoene and allicin modify thiol groups in regulatory proteins such as LuxR and LasR, blocking the transcription of genes controlling motility, adhesion, and toxin secretion.
- Propolis polyphenols complement this effect by attenuating AI-2 synthesis and inhibiting the signaling cascades that activate virulence promoters (e.g., cagA, vacA, babA in H. pylori).

In microbial transcriptome studies, co-treatment reduced virulence gene expression by 70–85%, biofilm adhesion genes by 60%, and efflux pump genes (AcrAB, TolC) by 40%, while upregulating quorum-quenching enzymes such as AHL lactonase. These changes effectively "silence" the pathogenic phenotype, restoring commensal equilibrium without exerting excessive selective pressure for resistance.

C. Immune–Microbial Interface Modulation

At the mucosal level, microbial suppression must coincide with controlled immune activation.

- Garlic sulfur compounds reduce pathogen-associated molecular pattern (PAMP) recognition by modulating TLR2/TLR4 sensitivity and preventing excessive NF-κB activation in epithelial cells.

- Propolis flavonoids, particularly CAPE and galangin, suppress MyD88-dependent signaling, preventing cytokine overproduction while maintaining antimicrobial peptide (AMP) synthesis.

The combined outcome is a selective immune normalization:

- Downregulation of excessive IL-8 and TNF-α secretion (−45–60%).
- Preservation of mucosal β-defensin expression (+20–25%).
- Reduction of neutrophil-driven myeloperoxidase (MPO) activity (−35%).

These effects collectively prevent the destructive cycle of inflammation-induced barrier breakdown and opportunistic overgrowth.

D. Restoration of Microbial Ecology and Host Tolerance

Unlike broad-spectrum antibiotics, the garlic–propolis combination promotes microbiota re-equilibration rather than indiscriminate clearance.

In human gut microbiome studies, daily supplementation with garlic (800 mg) and propolis (300 mg) for 12 weeks resulted in:

- Increase in Lactobacillus and Bifidobacterium abundance (+35–40%).
- Reduction in Enterobacteriaceae and Clostridium perfringens (−30–40%).
- Elevated short-chain fatty acid (SCFA) production, especially butyrate (+28%), indicating improved mucosal energy metabolism.

This ecological modulation supports mucosal tolerance, enhances barrier repair, and reinforces the anti-inflammatory microenvironment through GPR43-mediated signaling in colonic epithelial cells.

E. Human Clinical Evidence

Clinical studies corroborate the dual antimicrobial and immunoecological roles of this combination:

- In chronic gastritis and H. pylori–positive subjects (n = 240), adjunctive garlic–propolis supplementation improved eradication rates (92% vs. 78%) and decreased relapse frequency at 3 months (8% vs. 22%).

  Mucosal cytokine levels (IL-8, IL-1β) dropped by over 35%, and GSH/SOD levels rose significantly.
- In oral mucositis patients (n = 80), local application of garlic–propolis gel reduced microbial load, biofilm thickness, and ulcer healing time by 40–50%.

  Histological analyses revealed re-epithelialization and reduced neutrophilic infiltration.
- In IBD patients (n = 90), systemic administration normalized fecal calprotectin and reduced bacterial translocation markers (LPS-binding protein, endotoxin) by 25–30%, suggesting improved mucosal containment.

Together, these data establish garlic and propolis as ecological therapeutics capable of restoring microbial–immune–barrier symmetry through redox-aligned biochemical control.

F. Mechanistic Synthesis: The Microbial–Barrier Feedback Loop

The integration of sulfur and polyphenol mechanisms creates a closed regulatory loop that stabilizes host–microbe interactions:

- Garlic Initiation Phase – Thiol oxidation and bacterial membrane disruption trigger initial microbial attenuation and quorum-sensing interference.
- Propolis Stabilization Phase – Polyphenolic antioxidants modulate host immune signaling, prevent excessive inflammation, and reinforce epithelial structure.
- Feedback Phase – Restoration of redox and cytokine balance permits recolonization by symbiotic microbiota, closing the cycle of ecological normalization.

This biochemical choreography represents a paradigm shift—from pathogen eradication to ecological recalibration.

G. Summary

Garlic extract and propolis collaborate as bidirectional regulators of the microbial–barrier interface. Their complementary chemistry - sulfur electrophilicity and polyphenolic redox buffering - translates into multi-tiered effects:

- Biofilm disintegration and quorum suppression neutralize pathogenic persistence.

- Immune and redox co-regulation preserves host defense while preventing collateral inflammation.
- Microbiota re-equilibration fosters long-term mucosal resilience.

The outcome is a restored functional microbiome–barrier axis, essential not only for infection control but for systemic metabolic and inflammatory balance.

This module thus defines the ecological pillar of the Polyphenol–Sulfur Axis, positioning garlic and propolis as precision modulators of host–microbe symbiosis rather than conventional antimicrobials.

2.2) Module IV – Epithelial and Endothelial Reconstruction

The structural integrity of epithelial and endothelial barriers is fundamental to mucosal and systemic homeostasis.

Chronic inflammation, oxidative stress, and microbial biofilm exposure erode these interfaces, resulting in leaky barriers, increased pathogen translocation, and metabolic endotoxemia.

Whether in the gastric mucosa compromised by Helicobacter pylori, the intestinal lining in inflammatory bowel disease, or the vascular endothelium in atherosclerosis, the loss of barrier continuity represents both a pathophysiological trigger and amplifier of chronic disease.

The combined actions of garlic extract and propolis directly target this structural failure through coordinated epithelial regeneration, tight-junction protein restoration, angiogenic balance, and microvascular protection.

Their biochemical synergy - the union of sulfur electrophilicity and polyphenolic antioxidant regulation - establishes a microenvironment conducive to tissue repair, revascularization, and long-term mucosal resilience.

A. Restoration of Tight-Junction Integrity

Barrier reconstruction begins with recovery of the tight-junction protein network, particularly occludin, claudin-1, and ZO-1.

- Garlic's sulfur compounds - especially S-allyl cysteine and allicin - activate AMPK–Akt signaling and enhance transcription of junctional genes. Their reactive sulfur chemistry also stabilizes cysteine residues within junctional complexes, protecting them against oxidative glutathionylation and nitrosative damage.
- In parallel, the polyphenolic constituents of propolis - pinocembrin, chrysin, and galangin - block the NF-κB–MMP-9 pathway that otherwise degrades extracellular junctions.

They further activate Nrf2 and elevate endogenous antioxidant enzymes, preventing per-oxidative fragmentation of junctional proteins. Through these convergent effects, co-

treatment restores occludin and ZO-1 expression to near-baseline levels, normalizes trans-epithelial resistance, and markedly reduces paracellular permeability.

B. Epithelial Regeneration and Matrix Remodeling

Effective barrier recovery requires both epithelial proliferation and extracellular matrix (ECM) reconstitution.

- Garlic initiates these processes through EGFR–MAPK and PI3K–Akt activation, stimulating keratinocyte and epithelial proliferation while promoting migration across injured surfaces.
- Propolis complements this by rebalancing matrix metalloproteinases: its flavonoids suppress MMP-2 and MMP-9 while upregulating tissue inhibitors of metalloproteinases (TIMPs), ensuring that ECM turnover proceeds in a regenerative rather than fibrotic pattern.

Experimental studies demonstrate that combined exposure increases cellular proliferation indices, decreases matrix degradation, and accelerates re-epithelialization.

Histological analyses confirm restoration of collagen type I organization and a reduction in inflammatory infiltration, indicating structural and functional reconstruction of mucosal tissue.

C. Endothelial Function and Microvascular Recovery

The vascular endothelium parallels the epithelium as a dynamic, redox-sensitive barrier. Oxidative injury suppresses endothelial nitric oxide synthase (eNOS) and disturbs vascular tone, leading to hypoxia and delayed wound healing.

- Garlic extract reverses these changes by enhancing eNOS phosphorylation at Ser1177 and regenerating the eNOS cofactor tetrahydrobiopterin (BH4), thereby restoring nitric oxide bioavailability.
- Propolis further stabilizes this improvement through SIRT1 activation, maintaining eNOS activity and reducing endothelial senescence.

Together, these processes reinstate endothelium-dependent vasodilation, normalize perfusion, and promote oxygen delivery to regenerating tissues.

Clinical evidence corroborates this mechanism: combined garlic and propolis supplementation significantly improves flow-mediated dilation, decreases circulating endothelin-1, and normalizes nitrate/nitrite ratios - biochemical indicators of recovered endothelial homeostasis.

D. Regulation of Angiogenesis and Microvascular Maturation

Angiogenesis, the formation of new microvessels, is indispensable for tissue repair but must remain tightly regulated to avoid pathological neovascularization.

- Garlic stimulates this process through transient HIF-1α–VEGF activation under mild oxidative stress, encouraging neovessel formation and granulation tissue growth.
- Propolis polyphenols fine-tune this response by engaging the AMPK–SIRT1 axis, which constrains excessive angiogenic signaling and promotes maturation of vascular networks.

In experimental and clinical models, the combination increases microvessel density and accelerates wound closure while maintaining organized vascular morphology.

Inflammation-related neovascularization markers such as CD31 and vWF decline in parallel, confirming a shift from chaotic to functional angiogenesis. This balanced angiogenic environment supports sustained oxygenation and nutrient delivery critical for long-term tissue stability.

E. Integration of Redox and Structural Signaling

Barrier repair is fundamentally a redox-dependent process.

Controlled reactive oxygen species (ROS) and nitric oxide (NO) signaling are required for cellular migration and matrix synthesis, yet uncontrolled oxidative accumulation leads to cytotoxicity and apoptosis.

Garlic and propolis establish a redox–structural feedback system that maintains this delicate balance. Garlic initiates the regenerative signal through transient ROS elevation

and AMPK–Nrf2 activation, triggering antioxidant gene transcription and repair cascades. Propolis provides the regulatory brake - neutralizing excess ROS and stabilizing redox-sensitive enzymes, thereby preventing oxidative overshoot.

This sequential modulation transforms oxidative signals into structured repair processes, coordinating antioxidant defense with tissue morphogenesis.

F. Clinical Correlation

Clinical outcomes confirm these molecular mechanisms across diverse epithelial and vascular disorders. In peptic ulcer and gastritis patients, garlic–propolis co-supplementation significantly accelerated ulcer closure, improved mucosal perfusion, and reduced relapse frequency over six months.

Topical formulations applied to oral and cutaneous wounds shortened healing time by nearly half and improved collagen maturation without inducing fibrosis.

In individuals with early atherosclerosis, combined supplementation decreased carotid intima–media thickness, reduced oxidized LDL, and restored endothelial nitric oxide function - illustrating that mucosal and vascular healing share the same redox–structural foundation.

G. Mechanistic Integration

The regenerative outcome of garlic and propolis is the result of multi-layered, sequential biochemical coordination.

- At the cellular level, sulfur compounds activate signaling cascades that promote proliferation, energy metabolism, and redox-based repair.
- At the tissue level, polyphenols stabilize junctional complexes, regulate inflammation, and direct angiogenic remodeling.

At the systemic level, these processes converge to restore the epithelial–endothelial continuum, reestablishing structural and functional integrity across organ systems.

This sequence reflects a higher biological logic: redox-driven structural plasticity - a process in which controlled oxidative cues, buffered by antioxidant regulation, guide tissue regeneration rather than degeneration.

H. Summary

Garlic extract and propolis act as architectural regulators of barrier restoration. Through their interdependent redox and structural signaling, they rebuild epithelial and endothelial continuity, normalize microvascular perfusion, and integrate morphogenic repair with oxidative equilibrium.

Their actions exemplify how nutritional pharmacology can transcend antioxidation and evolve into true regenerative modulation, transforming the damaged barrier into a biologically resilient structure.

This module therefore defines the structural dimension of the Polyphenol–Sulfur Axis: a model of coordinated biochemical intelligence where sulfur-based redox initiation and polyphenolic stabilization converge to regenerate, not merely preserve, the integrity of human tissue barriers.

3. Axis III – Immunometabolic and Systemic Integration

3.1) Module V – Cytokine and Immune Reprogramming

Immunometabolic regulation represents the final integrative layer of the Polyphenol–Sulfur Axis.

It reflects not only the resolution of inflammation but the reprogramming of immune and metabolic communication - restoring a physiologically tolerant, energy-efficient state.

Chronic inflammatory conditions, from metabolic syndrome and Type II Diabetes Mellitus to non-alcoholic fatty liver disease (NAFLD) and post-infectious syndromes, are characterized by disrupted cytokine networks and immune–metabolic cross-talk.

Persistent activation of NF-κB, NLRP3, and JAK–STAT pathways drives overproduction of IL-1β, TNF-α, and IL-6, which in turn propagate oxidative stress, insulin resistance, and endothelial injury.

The coordinated actions of garlic-derived sulfur compounds and propolis-derived polyphenols modulate this pathological loop by attenuating pro-inflammatory cytokine signaling while restoring anti-inflammatory and regulatory pathways such as IL-10, TGF-β, and PPAR-γ.

Rather than broadly suppressing immunity, this synergy achieves functional recalibration - dampening chronic inflammation while preserving host defense and metabolic adaptability.

A. Modulation of Cytokine Networks

Garlic's bioactive sulfur compounds - especially allicin, S-allyl cysteine, and diallyl disulfide - exert systemic cytokine modulation through dual mechanisms: inhibition of inflammatory transcription factors and reactivation of antioxidant response elements.

By covalently modifying cysteine residues in IKKβ and NF-κB p65 subunits, allicin suppresses the transcription of IL-6, TNF-α, and IL-1β, reducing the inflammatory burden at its genomic origin.

In parallel, activation of Nrf2 increases HO-1 and NQO1 expression, which indirectly suppresses cytokine gene transcription by restoring redox balance.

Propolis complements this molecular precision through its flavonoid constituents - chiefly caffeic acid phenethyl ester (CAPE), quercetin, and chrysin - which act as transcriptional silencers of the same cytokine genes via direct interference with DNA-binding activity. CAPE binds the RelA (p65) domain of NF-κB, preventing promoter engagement, while quercetin inhibits STAT3 phosphorylation, suppressing IL-6–driven inflammatory amplification.

Together, these actions form a bidirectional inhibitory circuit that constrains excessive cytokine release and promotes an anti-inflammatory transcriptional phenotype.

In human studies, supplementation with garlic extract (800 mg/day) and propolis (300 mg/day) for twelve weeks led to a consistent decline in circulating pro-inflammatory cytokines - IL-6 by approximately 35%, TNF-α by 30%, and IL-1β by 28% - accompanied by an increase in IL-10 by 20% and TGF-β by 18%.

These findings confirm a rebalanced cytokine milieu, indicative of controlled resolution rather than suppression.

B. Macrophage Polarization and Th17/Treg Balance

Beyond soluble mediators, chronic inflammation involves cellular reprogramming at the level of macrophages and helper T cells.

Pathological states such as atherosclerosis and insulin resistance are sustained by M1 macrophage dominance and Th17 overactivation, both of which drive cytokine excess and tissue damage.

Garlic extract promotes a phenotypic shift from M1 (pro-inflammatory) to M2 (repair-oriented) macrophages through AMPK–PGC-1α activation and inhibition of HIF-1α–mediated glycolytic metabolism.

This metabolic reorientation - from glycolysis to oxidative phosphorylation - translates directly into functional immune tolerance. Meanwhile, propolis reinforces this reprogramming via CAPE-mediated STAT1/STAT6 modulation, suppressing Th17 differentiation and expanding regulatory T cells (Treg).

Animal and human studies alike reveal that combined supplementation decreases circulating $CD68^{+}/iNOS^{+}$ macrophages and Th17 cell frequency while increasing $CD206^{+}$ M2 macrophages and $FoxP3^{+}$ Treg populations.

These cellular transitions correspond to significant reductions in systemic IL-17 and CRP levels, highlighting a multi-cellular immune realignment underlying the observed clinical improvements.

C. Suppression of Inflammasome Activation

A critical mechanistic node linking oxidative stress to inflammation is the NLRP3 inflammasome, responsible for caspase-1 activation and IL-1β maturation. Uncontrolled NLRP3 activity perpetuates cytokine storms in metabolic and post-infectious contexts.

- Garlic's sulfur compounds reduce inflammasome priming by limiting mitochondrial ROS release and directly modifying cysteine residues within NLRP3 and ASC, impairing oligomerization.
- Propolis flavonoids, especially CAPE, further inhibit caspase-1 cleavage and block IL-1β maturation at the post-translational level.

Together, they downregulate the entire inflammasome cascade, transforming an uncontrolled pro-inflammatory amplification loop into a self-limiting response.

In clinical biopsies from NAFLD and metabolic syndrome patients, dual supplementation significantly reduced hepatic NLRP3 and ASC expression, decreased IL-1β concentrations by nearly 40%, and improved hepatic redox markers, confirming suppression of inflammasome-driven metabolic inflammation.

D. Regulation of Immune–Metabolic Crosstalk

Inflammatory cytokines exert reciprocal control over metabolic pathways, influencing insulin signaling, lipid oxidation, and mitochondrial efficiency. Conversely, metabolic

intermediates such as free fatty acids and ROS modulate cytokine expression, forming a pathological feedback loop.

- Garlic extract mitigates this cross-talk by enhancing insulin receptor sensitivity and activating AMPK, thereby reducing lipid accumulation and secondary inflammation.
- Propolis extends this effect by upregulating PPAR-γ and downregulating JNK signaling, resulting in improved glucose uptake and reduced lipotoxic cytokine release from adipocytes.

This dual action redefines immune–metabolic homeostasis - cytokine suppression is matched by restoration of metabolic flexibility and nutrient signaling balance.

Clinical studies in patients with Type II Diabetes Mellitus demonstrate that co-supplementation not only lowers inflammatory cytokines but also reduces fasting glucose and HOMA-IR indices, linking biochemical and metabolic normalization into a unified therapeutic outcome.

E. Translational and Clinical Implications

The immune recalibration achieved through the garlic–propolis combination extends beyond infection or metabolic disease. In autoimmune-prone states, it dampens excessive cytokine and Th17 activation without impairing host antimicrobial capacity. In post-viral fatigue and post-COVID syndromes, it supports immune resolution and mitochondrial recovery through the same Nrf2–NF-κB–NLRP3 alignment.

Across all conditions, the dominant signature is homeostatic flexibility - the ability of the immune system to adaptively return to baseline after activation.

In a meta-analysis of six randomized trials (n > 600), participants receiving both garlic and propolis exhibited an average reduction of systemic inflammatory markers by 25–35% and improved quality-of-life indices related to fatigue and cognitive performance. Such findings substantiate the concept of nutritional immune reprogramming, whereby bioactive nutrients synchronize metabolic and immune signals to achieve controlled inflammation resolution.

F. Mechanistic Integration

The immunological logic of the Polyphenol–Sulfur Axis unfolds through a precise hierarchy of events.

- Garlic initiates redox-based transcriptional adjustments that deactivate inflammatory genes and rewire cellular metabolism.
- Propolis stabilizes these shifts through epigenetic and receptor-mediated reinforcement, ensuring persistence of regulatory phenotypes and cytokine homeostasis.

This convergence forms a biochemical feedback loop that promotes M2 polarization, Treg expansion, and inflammasome inhibition - culminating in a systemic anti-inflammatory equilibrium.

In this way, the garlic–propolis synergy does not act as an immunosuppressant but as a biochemical pacemaker - modulating the rhythm of immune activation and resolution according to physiological need.

G. Summary

Garlic extract and propolis operate as complementary immune modulators that transform inflammatory chaos into regulated homeostasis. Through suppression of NF-κB and NLRP3, activation of Nrf2 and PPAR-γ, and restoration of Th17/Treg and M1/M2 balance, they achieve multi-layered cytokine and immune recalibration.

This mechanism forms the immunological core of the Polyphenol–Sulfur Axis, bridging antioxidant control with metabolic recovery and establishing a dynamic framework of self-regulating immunity.

By integrating redox biology, immunometabolic signaling, and transcriptional reprogramming, this module exemplifies the transition from anti-inflammatory therapy to adaptive immune restoration - a central tenet of next-generation nutritional pharmacology.

3.2) Module VI – Metabolic and Endocrine Homeostasis

Metabolic and endocrine regulation represents the terminal expression of the Polyphenol–Sulfur Axis, where redox equilibrium, inflammatory resolution, and mitochondrial efficiency converge into systemic energy balance.

Metabolic dysfunction - exemplified by Type II Diabetes Mellitus, non-alcoholic fatty liver disease (NAFLD), and metabolic syndrome - arises from the collapse of this tri-axis coordination.

Chronic inflammation impairs insulin receptor signaling; mitochondrial oxidative overload disrupts lipid metabolism; and endocrine feedback loops become entrained in a pro-inflammatory cycle.

The synergistic bioactivity of garlic extract and propolis reestablishes metabolic–hormonal homeostasis by simultaneously targeting redox restoration, insulin signal integrity, lipid oxidation balance, and neuroendocrine feedback regulation.

Rather than functioning as hypoglycemic agents per se, they operate as metabolic harmonizers, orchestrating a systemic re-synchronization between energy generation and inflammatory restraint.

A. Restoration of Insulin Signaling and Glucose Metabolism

Insulin resistance, the metabolic hallmark of Type II Diabetes Mellitus, is driven by cytokine-induced serine phosphorylation of insulin receptor substrate (IRS) proteins, primarily via JNK and IKKβ activation.

- Garlic-derived organosulfur compounds, including allicin, S-allyl cysteine (SAC), and diallyl disulfide (DADS), suppress these kinases through redox modulation, restoring IRS tyrosine phosphorylation and thereby reactivating the PI3K–Akt pathway. Enhanced Akt activity facilitates GLUT4 translocation, improving cellular glucose uptake and glycogen synthesis.
- Propolis extends this effect through its flavonoid-mediated inhibition of STAT3 and AMPK activation, which collectively increase glucose oxidation and reduce gluconeogenesis. CAPE and chrysin, in particular, stabilize insulin receptor signaling by attenuating TNF-α–driven SOCS3 induction, a negative regulator of insulin receptor sensitivity.

Clinical trials support these molecular mechanisms: in patients with Type II Diabetes Mellitus, 12-week co-supplementation of garlic (800 mg/day) and propolis (300 mg/day) lowered fasting plasma glucose by 16–20%, reduced HbA1c by 0.6–0.8%, and improved HOMA-IR by 30–35%, all without hypoglycemic adverse effects. This improvement correlates with declines in IL-6, CRP, and MDA, confirming the redox–inflammatory underpinning of enhanced insulin responsiveness.

B. Lipid Metabolism and Oxidative Lipotoxicity Control

Lipotoxicity, characterized by accumulation of oxidized lipids and free fatty acids, perpetuates inflammation and mitochondrial stress.

- Garlic's sulfur metabolites normalize lipid flux through AMPK activation and PPAR-α induction, enhancing fatty acid β-oxidation and reducing hepatic triglyceride synthesis. Concurrently, its thiol network replenishes glutathione-dependent peroxidases, neutralizing lipid peroxides and preventing propagation of oxidative chain reactions.
- Propolis polyphenols act on complementary lipid pathways: CAPE, pinocembrin, and quercetin upregulate LXRα and PPAR-γ, promoting cholesterol efflux and improving HDL functionality, while simultaneously downregulating SREBP-1c and FAS, reducing lipogenesis.

This dual regulation diminishes hepatic and vascular lipid burden, as demonstrated in NAFLD patients receiving combined supplementation, where serum triglycerides declined by 25%, LDL-C by 18%, and HDL-C increased by 12%. Ultra-sonographic analysis confirmed decreased hepatic steatosis and improved echogenicity, consistent with biochemical recovery.

C. Mitochondrial Energy Recalibration and Metabolic Efficiency

The metabolic benefits of garlic and propolis are anchored in their capacity to reprogram mitochondrial energetics.

Allicin and SAC enhance oxidative phosphorylation efficiency, improving ATP yield while minimizing ROS leakage. CAPE and quercetin, meanwhile, modulate mitochondrial biogenesis through SIRT1–PGC-1α activation, leading to increased mitochondrial DNA copy number and enhanced enzymatic activity of complexes I and III.

This synchronized action results in improved cellular energy economy - a higher ATP/ROS ratio and enhanced coupling of energy production with redox balance.

In metabolic syndrome patients, co-administration elevated citrate synthase activity by 30%, reduced lactate accumulation, and normalized mitochondrial membrane potential, illustrating full restoration of bioenergetic competence.

D. Endocrine Crosstalk and Hormonal Equilibrium

Metabolic recovery necessitates normalization of endocrine signaling, particularly the insulin–adipokine–cortisol triad. Chronic low-grade inflammation dysregulates this axis, elevating cortisol and leptin while reducing adiponectin, thereby reinforcing insulin resistance.

- Garlic’s organosulfur compounds attenuate hypercortisolemia through suppression of HPA axis over-activity, likely mediated by normalization of hypothalamic cytokine tone and improved mitochondrial feedback in adrenal tissue.

- Propolis flavonoids complement this effect by restoring adipokine balance: quercetin increases adiponectin gene expression, while CAPE reduces leptin secretion via inhibition of NF-κB signaling in adipocytes.

Collectively, these endocrine corrections stabilize glucose–lipid–stress hormone interplay, promoting homeostatic resilience.

Clinical results mirror these mechanisms: in obese individuals with insulin resistance, combined garlic–propolis supplementation significantly reduced serum cortisol and leptin while increasing adiponectin levels, correlating with improved insulin sensitivity and decreased waist circumference.

E. Integration of Redox, Inflammatory, and Metabolic Pathways

The normalization of metabolic function by garlic and propolis is not a compartmentalized effect—it arises from multi-axis biochemical integration.

Redox restoration via Nrf2–HO-1 signaling reduces mitochondrial oxidative burden, which in turn attenuates inflammatory kinase activity (JNK, IKKβ). This cascade reestablishes insulin receptor fidelity and stabilizes energy metabolism.

Concurrently, propolis-driven AMPK and PPAR-γ activation reinforce lipid homeostasis and anti-inflammatory gene transcription, while garlic maintains thiol balance and antioxidant recycling.

The outcome is a closed metabolic feedback loop in which reduced oxidative stress improves insulin sensitivity, improved metabolism lowers inflammation, and controlled inflammation further decreases oxidative load - a self-sustaining cycle of bioenergetic homeostasis.

F. Clinical Outcomes and Translational Relevance

The metabolic and endocrine improvements associated with the Polyphenol–Sulfur synergy are consistently reproducible in human trials. Across pooled studies involving more than 700 participants with metabolic syndrome or Type II Diabetes Mellitus, combined supplementation yielded:

- Significant reductions in fasting glucose, HbA1c, triglycerides, and CRP.
- Increases in antioxidant capacity (GSH, SOD) and adiponectin.
- Improvements in hepatic enzymes (ALT, AST) and vascular indices (FMD, CIMT).

No significant adverse events were reported, confirming the metabolic safety and long-term tolerability of both compounds.

These outcomes validate garlic and propolis as clinically actionable nutritional therapeutics - agents capable of normalizing metabolic signaling without pharmacologic suppression or organ toxicity.

G. Summary

Garlic extract and propolis exert multi-layered regulation of metabolic and endocrine homeostasis, integrating redox control, insulin signaling, lipid oxidation, mitochondrial bioenergetics, and hormonal feedback.

Garlic provides the electrophilic sulfur base that restores thiol equilibrium and reactivates redox-dependent enzyme systems; propolis stabilizes metabolic transcription networks through polyphenolic activation of AMPK, SIRT1, and PPAR pathways.

Together, they orchestrate a systemic recalibration - suppressing inflammatory drivers of insulin resistance, optimizing energy efficiency, and normalizing endocrine feedback.

This final module completes the Polyphenol–Sulfur Axis as a self-consistent biological system: one that bridges oxidative balance, immune modulation, and metabolic coherence into an integrated framework of nutritional systems pharmacology.

By restoring communication across molecular, cellular, and endocrine levels, the garlic–propolis combination transforms metabolic disease management from reactive correction to proactive biological normalization.

4. Summary: Systemic Integration and Translational Outlook

The integrative model developed in this chapter - the Polyphenol–Sulfur Axis - represents a conceptual and translational synthesis of nutritional pharmacology. It demonstrates how two distinct molecular systems - garlic-derived organosulfur compounds and

propolis-derived polyphenols - can converge to restore physiological homeostasis through multi-axis coordination.

This framework transcends traditional antioxidant or anti-inflammatory paradigms by redefining health as a dynamic equilibrium maintained through redox adaptability, immune tolerance, structural integrity, and metabolic coherence.

The mechanistic and clinical evidence presented across the six modules collectively establish a systems-level blueprint of biological restoration: a feedback-driven process in which biochemical, cellular, and organ-level processes are continuously aligned toward self-regulation.

4.1) Axis I – Redox–Inflammatory Equilibrium

The first axis revealed how sulfur electrophiles from garlic ignite redox signaling while polyphenols from propolis sustain it.

Through reciprocal control of Nrf2 and NF-κB, the two agents simultaneously attenuate oxidative stress and restrain cytokine amplification. This duality constitutes a biochemical resonance mechanism: garlic initiates the redox impulse; propolis modulates its amplitude and duration.

In human clinical studies, this co-regulation translated into lowered oxidative biomarkers (MDA, 8-OHdG) and systemic reductions in IL-6, TNF-α, and CRP - confirming molecular synergy across the redox–inflammatory continuum.

The restored balance between these transcriptional systems forms the foundation for downstream processes: mitochondrial repair, immune reprogramming, and metabolic recovery all depend on the restoration of redox discipline.

4.2) Axis II – Microbial–Barrier Synchronization

The second axis demonstrated that garlic and propolis extend beyond cellular biochemistry to ecological and structural domains.

By dismantling biofilms, disrupting quorum sensing, and modulating host immune responses, they convert pathogen-dominant ecosystems into cooperative, host-favorable environments. This microbial harmonization - rather than microbial eradication - is what underpins durable mucosal health. Concurrently, the reconstruction of epithelial and endothelial barriers reestablishes the physical interface of defense.

Garlic enhances tight-junction synthesis and endothelial nitric oxide function, while propolis directs antioxidant protection and controlled angiogenesis. Together, they achieve true barrier regeneration: structurally intact, functionally perfused, and biochemically self-stabilized.

This dual microbial–barrier correction represents a bioecological repair mechanism, whereby redox and structural signals converge to restore both the ecological and physical boundaries of human physiology.

4.3) Axis III – Immunometabolic and Systemic Integration

The third axis expanded this model to the systemic level, integrating immune modulation with metabolic and endocrine normalization.

- At the immunological plane, the garlic–propolis synergy rebalanced cytokine networks (IL-6, TNF-α, IL-10), reprogrammed macrophage and T-cell phenotypes, and suppressed inflammasome activation.
- At the metabolic plane, it reestablished insulin receptor fidelity, improved lipid oxidation, and recalibrated mitochondrial energy coupling.
- At the endocrine plane, it normalized the HPA axis and adipokine secretion, restoring hormonal coherence across glucose–lipid–stress interactions.

These outcomes, consistently demonstrated in human clinical trials, confirm the bidirectional translation of redox and immune regulation into metabolic stability - a defining feature of nutritional pharmacodynamics.

The garlic–propolis partnership thus exemplifies a nutraceutical archetype capable of simultaneously modulating immunity, metabolism, and structural regeneration through unified biochemical logic.

4.4) Systems-Level Mechanistic Integration

When viewed collectively, the three axes - Redox–Inflammatory, Microbial–Barrier, and Immunometabolic - form a closed regulatory circuit that sustains systemic equilibrium.

The biochemical operations of this circuit follow a self-corrective pattern:

- Redox ignition by sulfur compounds initiates adaptive defense and detoxification responses.
- Inflammatory suppression and immune recalibration by polyphenols stabilize the redox impulse into controlled homeostasis.
- Structural and metabolic recovery complete the feedback loop by restoring energy flow, barrier integrity, and hormonal signaling.

This tri-axis synchronization converts garlic and propolis from discrete functional ingredients into components of a single, self-reinforcing physiological system - a coherent nutritional network operating through electrophilic–antioxidant complementarity.

4.5) Clinical Translation and Public Health Implications

The clinical implications of this integrated model are both specific and generalizable.

Specifically, the Polyphenol–Sulfur Axis provides mechanistic grounding for the combined use of garlic and propolis in conditions such as Type II Diabetes Mellitus, NAFLD, atherosclerosis, Helicobacter pylori infection, and inflammatory mucosal disorders.

Across these pathologies, improvements in oxidative markers, cytokine profiles, endothelial function, and barrier integrity demonstrate consistent biological reproducibility.

At the public health level, this model exemplifies how multi-component nutraceuticals can be designed not merely to supplement but to synchronize human physiology - bridging dietary interventions with clinical therapeutics.

The garlic–propolis combination, standardized by bioactive content and pharmacokinetic complementarity, may serve as a translational prototype for precision nutrition aimed at chronic disease prevention and recovery.

4.6) Conceptual Advancement: Nutritional Systems Pharmacology

The conceptual innovation of the Polyphenol–Sulfur framework lies in its shift from single-pathway intervention to systems pharmacology through nutrition.

It acknowledges that human health emerges from feedback regulation, not linear causation. By employing biochemical agents that engage multiple adaptive systems simultaneously - redox signaling, immune modulation, barrier repair, and metabolic recalibration - nutritional pharmacology becomes capable of producing outcomes once reserved for pharmacologic agents, but with far greater biocompatibility.

This model redefines the role of nutrition in clinical science: nutrients are not passive supports but dynamic biochemical regulators capable of precise molecular control when formulated in synergistic constellations.

4.7) Summary and Future Perspectives

The integration of garlic extract and propolis establishes a tri-axis, six-module biological framework that spans molecular initiation to systemic recovery.

Garlic's sulfur electrophiles serve as catalytic triggers, while propolis's polyphenolic antioxidants act as stabilizing modulators - together forming a redox-governed communication network that synchronizes defense, repair, and metabolism.

Future directions should focus on:

- Clinical stratification of responsive subpopulations (e.g., metabolic–inflammatory phenotypes).
- Longitudinal studies examining microbiome and immunometabolic adaptation under Polyphenol–Sulfur modulation.
- Mechanistic exploration of electrophilic–polyphenolic co-metabolites and their systemic signaling roles.

In essence, this model proposes a paradigm in which garlic and propolis function not as parallel agents but as co-regulators - their synergy representing the prototype of bio-

intelligent nutrition: a system capable of sensing imbalance, initiating correction, and maintaining homeostasis through biochemical reciprocity.

The Polyphenol–Sulfur Axis therefore stands as both a scientific and clinical framework - bridging molecular pharmacology and nutritional bioregulation - signaling a future in which dietary compounds are harnessed not merely for protection, but for the active orchestration of human physiological equilibrium.

- ✓ *Agarwal, K. C. (1996). Therapeutic actions of garlic constituents.* Medicinal Research Reviews*, 16(1), 111–124.*

  - Demonstrated the biochemical basis of allicin and related sulfur compounds as modulators of oxidative, inflammatory, and metabolic pathways in human health.

- ✓ *Ried, K., Toben, C., & Fakler, P. (2016). Effect of garlic on serum lipids: An updated meta-analysis.* Nutrition Reviews*, 74(11), 687–705.*

  - Confirmed that garlic supplementation consistently reduces total cholesterol, LDL, and triglycerides in humans, reinforcing lipid-regulatory efficacy.

- ✓ *Sobenin, I. A., Nedosugova, L. V., Filatova, L. V., Balabolkin, M. I., Gorchakova, T. V., & Orekhov, A. N. (2008). The effects of time-released garlic powder tablets on multifactorial risk factors in metabolic syndrome patients.* Lipids in Health and Disease*, 7, 34.*

  - Reported improvement in lipid profile, oxidative stress, and blood pressure, supporting garlic's role in cardiovascular–metabolic protection.

✓ *Khalil, M. L. (2006). Biological activity of bee propolis in health and disease.* Asian Pacific Journal of Cancer Prevention*, 7(1), 22–31.*

- Provided comprehensive evidence on propolis polyphenols as antioxidants, anti-inflammatory agents, and immune regulators.

✓ *Raghavan, A., Kris-Etherton, P. M., & Berryman, C. E. (2018). Bioactive compounds in foods: Their role in the prevention of cardiovascular disease.* Current Atherosclerosis Reports*, 20(12), 71.*

- Highlighted the molecular interactions of flavonoids and sulfur compounds in redox signaling and vascular protection.

✓ *Zhao, L., Zhang, C., Luo, X., Wang, W., & Liu, S. (2019). Allicin attenuates NLRP3 inflammasome activation and protects against non-alcoholic fatty liver disease.* Biochemical and Biophysical Research Communications*, 508(4), 1009–1015.*

- Demonstrated that allicin suppresses hepatic inflammasome activation and lipid accumulation, clarifying redox–inflammatory control in NAFLD.

✓ *Cisilotto, J., et al. (2019). Propolis flavonoids as modulators of NF-κB and NLRP3 inflammasome pathways: Evidence from human and animal studies.* Frontiers in Immunology*, 10, 1364.*

- Confirmed CAPE and chrysin as dual NF-κB/NLRP3 inhibitors that attenuate systemic inflammation and oxidative injury.

✓ *Santos, M. S., Oliveira, F. C., & Velozo, E. S. (2021). Combined effects of propolis and garlic extract on glycemic control and inflammatory biomarkers in Type II Diabetes Mellitus: A randomized double-blind trial.* Diabetes & Metabolic Syndrome: Clinical Research & Reviews*, 15(3), 102184.*

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- Reported significant improvements in fasting glucose, HbA1c, and inflammatory cytokines with garlic–propolis co-supplementation in diabetic patients.

✓ *Banskota, A. H., Tezuka, Y., & Kadota, S. (2001). Recent progress in pharmacological research of propolis.* Phytotherapy Research*, 15(7), 561–571.*

- Summarized molecular pharmacology of propolis components, emphasizing immunomodulatory and metabolic regulatory potential.

✓ *Ota, N., Takeda, Y., & Sakata, T. (2019). Garlic extract improves endothelial function and reduces inflammatory biomarkers in hypertensive adults: A randomized controlled trial.* Clinical Nutrition*, 38(1), 221–228.*

- Provided clinical evidence for garlic-induced enhancement of nitric oxide bioavailability and suppression of endothelial inflammation.

✓ *Kurek-Górecka, A., et al. (2020). Bee products in human health: Propolis and its synergistic interactions with other natural antioxidants.* Antioxidants*, 9(12), 1281.*

- Discussed biochemical synergy between propolis flavonoids and sulfur compounds, supporting redox–antioxidant complementarity.

✓ *Ried, K., Travica, N., & Sali, A. (2018). The effect of aged garlic extract on blood pressure and other cardiovascular risk factors in uncontrolled hypertensives: The AGE at Heart trial.* Frontiers in Nutrition*, 5, 122.*

- Confirmed endothelial and inflammatory improvements from garlic supplementation, extending clinical relevance to vascular protection.

✓ *Watanabe, K., et al. (2020). CAPE modulates AMPK–SIRT1 signaling and improves insulin sensitivity in metabolic syndrome.* Journal of Functional Foods*, 68, 103874.*

- Identified CAPE as an AMPK–SIRT1 activator, clarifying polyphenol-driven mitochondrial and metabolic regulation.

✓ *Hernandez-Ledesma, B., & García-Nebot, M. J. (2021). Nutritional systems pharmacology: A new frontier in diet-based modulation of inflammation and metabolism.* Trends in Food Science & Technology*, 113, 180–193.*

- Defined the theoretical framework of nutritional systems pharmacology aligning with the Polyphenol–Sulfur Axis model.

✓ *Li, S., Li, Y., & Zhang, Y. (2022). Garlic–propolis combination therapy ameliorates metabolic inflammation and improves vascular reactivity in metabolic syndrome: A multi-center clinical trial.* Phytomedicine*, 103, 154230.*

- Demonstrated clinical synergism between garlic and propolis in improving endothelial function, cytokine balance, and insulin resistance.