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Sourdough Bioactives from Fermentation to Human Physiology: Biosynthesis, Bioavailability, Mechanisms and Clinical Evidence

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ABSTRACT
Sourdough is a metabolically active cereal ecosystem in which lactic acid bacteria, yeasts and endogenous flour enzymes transform carbohydrates, proteins and phytochemicals. These reactions can increase organic acids, release phenolic compounds, generate peptides, form exopolysaccharides and, in strain- tand substrate-dependent systems, enrich γ-aminobutyric acid and selected vitamins. However, the detection of a compound in fermented dough does not establish physiological benefit: This narrative review organizes the evidence from precursor and microbial pathway to processing stability, gastrointestinal fate, mechanical and clinical effect. Appraisal was narrative, not a systematic review; evidence strength reflected design limitations, consistency, directness, precision and plausibility. Strong evidence supports acidification, phytate degradation, phenolic release and technological functions of exopolysaccharides. Mechanistic evidence for antioxidant, antihypertensive, immunomodulatory and microbiota-mediated effects is plausible but is dominated by chemical assays, cell models and studies of purified compounds. Human trials mainly examine postprandial glycaemia, gastrointestinal responses and satiety; their results are heterogeneous and do not establish that sourdough fermentation alone prevents chronic disease. Flour composition, strain identity, fermentation time, temperature, hydration and baking interact, making the final bread not the fermented dough the relevant exposure. Future work should employ chemically characterized breads, matched comparators, simulated digestion linked to targeted metabolomics, dose–response designs and adequately powered randomized trials. Sourdough is best viewed as a controllable biotransformation platform whose health value depends on the product-specific survival and bioavailability of its metabolites rather than on fermentation as a generic label.

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INTRODUCTION

Sourdough is produced by fermenting flour and water with a microbial consortium dominated by lactic acid bacteria (LAB) and yeasts. Its acidity, aroma and leavening arise from a network of carbohydrate utilization, amino-acid conversion, redox balancing and cross-feeding rather than from a single organism. This ecological complexity explains both sourdough's technological versatility and a central problem in health research: two products described as sourdough may differ markedly in flour, microbial composition, fermentation intensity and final metabolite profile (Gänzle & Ripari, 2016; De Vuyst *et al*., 2017; Siepmann *et al*., 2018).

Interest has shifted from sensory quality and preservation toward the possibility that sourdough functions as a food biotransformation system. Fermentation can acidify the cereal matrix, activate endogenous enzymes, increase proteolysis, release cell-wall-bound phenolics, reduce fermentable carbohydrates and phytate, and generate microbial polymers or metabolites. These effects provide credible routes to changes in mineral bioaccessibility, starch digestion, gastrointestinal tolerance and the chemical antioxidant capacity of bread (Arora *et al*., 2021a, 2021b; De Bondt *et al*., 2024; Pérez-Alvarado *et al*., 2022). Yet the term bioactive is often applied too readily. A higher DPPH value in a dough extract, for example, is not equivalent to improved antioxidant status in humans, and ACE inhibition by an undigested peptide extract does not show a blood-pressure effect after bread consumption.

The distinction between formation and function is especially important because baking transforms the exposure. Heat can degrade vitamins and peptides, alter phenolic extractability, depolymerize or cross-link macromolecules and produce new Maillard-reaction products. Digestion subsequently changes pH, enzymatically hydrolyses proteins and polysaccharides, and exposes metabolites to absorption or colonic fermentation (Landis *et al*., 2021). Consequently, the correct unit of inference is the chemically characterized final bread together with its gastrointestinal fate, not the fermented dough alone.

Recent systematic evidence reinforces this caution. A review of 25 clinical trials involving 542 participants found that sourdough products sometimes improved glycaemic response, satiety or gastrointestinal comfort, but heterogeneity in cereal type, starter, fermentation and comparator prevented a general conclusion that sourdough per se is healthier than yeast-leavened bread (Ribet *et al*., 2023). A randomized crossover study likewise showed pronounced interpersonal glycaemic responses and no uniform superiority of one bread type, illustrating the importance of flour composition, host phenotype and microbiome context (Korem *et al*., 2017).

This review addresses the resulting translational gap. It evaluates the evidence chain cereal precursor. Microorganism

and enzyme to the quantity retained in bread, its bioaccessibility, mechanistic plausibility and human outcome. The review defines the major sourdough derived bioactive classes, explains the microbial and biochemical routes governing their production, and evaluate processing stability and gastrointestinal fate. It distinguishes direct sourdough evidence extrapolated from purified compounds or other fermented foods and identifies experiments required to convert promising mechanisms into defensible health claims. The aim is to critically evaluate the evidence linking sourdough bioactive formation to human physiological effects.

MATERIALS AND METHODS

Search Reproducibility and Study Selection

This narrative review used a structured, topic-based literature search updated on 3 September 2026. PubMed was searched from database inception using combinations of the following concepts: (sourdough OR sourdough bread), (lactic acid bacteria OR yeast OR microbiome OR fermentation), (bioactive OR phenolic OR peptide OR GABA OR exopolysaccharide OR vitamin OR phytate OR mineral), (digestion OR bioaccessibility OR bioavailability), and (glycaemic OR gastrointestinal OR clinical trial OR metabolomics OR metagenomics). Boolean combinations were adapted to each topic rather than treated as one indiscriminate query. Reference lists of eligible reviews and key primary studies were screened backward, and recent records were checked against PubMed metadata and DOI information. Because the aim was critical mechanistic integration rather than estimation of a pooled effect, the review was not registered and no meta-analysis was performed. The resulting conclusions should therefore be interpreted as a structured critical synthesis, not as a PRISMA-compliant systematic review.

Eligible publications were peer-reviewed studies in English that examined sourdough microbial ecology; formation or transformation of bioactives; retention during baking or storage; gastrointestinal fate; cellular, animal or human mechanisms; or clinical outcomes. Human evidence required ingestion of a chemically or technologically described sourdough product. Studies of fermented foods generally, purified compounds, simulated digestion, cell systems and animal models were retained only as indirect mechanistic evidence and were labelled accordingly. Editorials, non-peer-reviewed reports, studies lacking an identifiable fermentation treatment and papers making health claims without an analytical or biological outcome were excluded.

Data were extracted for cereal species and extraction rate, starter identity, inoculum, dough yield, fermentation time and temperature, pH or total titratable acidity, baking and storage conditions, analytical platform, comparator, sample size, population, intervention duration, outcomes and author-reported limitations. Human studies were appraised across randomization, blinding, allocation concealment, comparator matching, carry-over and washout, attrition, selective reporting, product characterization and control of dietary confounding. Certainty was assigned as high, moderate, low or very low by considering design limitations, consistency, directness, precision and plausibility; mechanistic evidence could support interpretation but could not upgrade an unsupported clinical claim.

Evidence was interpreted across five levels. Level 1 comprises compositional or chemical-assay evidence; Level 2 comprises simulated digestion and cell or ex vivo models; Level 3 comprises animal studies; Level 4 comprises acute or short-term human feeding studies; and Level 5 comprises controlled human interventions with clinically relevant endpoints. Confidence increases when the tested exposure is a chemically characterized sourdough bread, the comparator is matched for flour and processing, metabolite retention is quantified, and outcomes are replicated. Confidence decreases when results are extrapolated from purified compounds, predicted glycaemic index, nonspecific antioxidant assays or fermented foods unrelated to sourdough.

Table 1: Translational Evidence Hierarchy and Permissible Inference Limits

Evidence Level	Scope and Methodologies	Permissible Scientific Inference	References
Composition and chemical assays	Compound identification; peptide sequencing; radical-scavenging assays; enzyme-inhibition assays.	Establishes chemical composition and activity under assay conditions; does not demonstrate physiological benefit in humans.	Boudaoud et al. (2021); Korcari et al. (2021); Brand and Wefers (2025b).
Digestion, transport and cell models	Simulated gastrointestinal digestion; epithelial transport models; cellular-response assays.	Assesses bioaccessibility, digestive stability and transport potential under experimental conditions; supports mechanistic interpretation. Systemic bioavailability requires in vivo uptake or exposure evidence.	Coda et al. (2012); Liu et al. (2018); Yu et al. (2024).
Preclinical animal models	Controlled feeding interventions; metabolite tracking; physiological and tissue assessments.	Supports biological plausibility and model-specific physiological or toxicity findings; does not establish efficacy or safety in humans.	Yu et al. (2024); Kim et al. (2025).
Acute or short-term human feeding studies	Controlled meal comparisons; glucose and insulin measurements; appetite ratings; gastrointestinal-response assessments.	Identifies immediate or short-term responses to the tested product; does not establish sustained benefit or chronic disease prevention.	Korem et al. (2017); Zamaratskaia et al. (2017); Iversen et al. (2018).
Controlled human interventions with clinically relevant endpoints	Repeated-consumption trials; matched comparators; validated clinical outcomes; adverse-event monitoring.	Evaluates effects within the tested population, dose and duration. Product-specific health claims require adequate trial quality, precision and supporting evidence; broader benefits cannot be assumed.	Laatikainen et al. (2017); Rizzello et al. (2019); Pérez-Vega et al. (2024).

RESULTS AND DISCUSSION

Ecological assembly occurs at several scales. Flour introduces microorganisms and substrates, the bakery environment and handling practices contribute dispersal, and repeated refreshments select organisms able to tolerate acidification, maltose competition and recurrent dilution. Consequently, geographically distinct starters can converge functionally while retaining taxonomic differences, and apparently similar starters can diverge when flour or feeding schedules change (Oshiro *et al*., 2021; Gänzle *et al*., 2023; Zhour *et al*., 2026).

Functional annotation should therefore move below the species level. Starter selection benefits from strain-resolved genomics combined with phenotype measurements for acidification, proteolysis, phenolic conversion, EPS synthesis and antifungal activity. Synthetic-community and perturbation experiments are especially valuable because they can separate correlation from cross-feeding, competition and metabolic complementation (De Vuyst *et al*., 2009; McKenney *et al*., 2023; Mietton *et al*., 2024; Franco *et al*., 2020; Nissen *et al*., 2024).

Mature type I sourdoughs commonly contain acid-adapted LAB including *Fructilactobacillus sanfranciscensis*, *Lactiplantibacillus plantarum*, *Levilactobacillus brevis* and *Limosilactobacillus* fermentum and yeasts such as *Saccharomyces cerevisiae* and *Kazachstania humilis*. Their relative abundance varies with geography, flour, temperature, dough yield and propagation regime. Modern taxonomic nomenclature should be used consistently because the former genus Lactobacillus was reorganized into multiple genera (Zheng *et al*., 2020).

Species lists alone poorly predict function. Strains of the same species may differ in proteolysis, phenolic-acid metabolism, glutamate decarboxylation, fructan utilization or exopolysaccharide production. Community function also emerges from cross-feeding: yeasts may provide amino acids and vitamins, LAB can hydrolyse cereal substrates and acidification changes both microbial competition and endogenous flour-enzyme activity. Functional redundancy can stabilize acidification while still allowing major differences in peptide, volatile and phenolic profiles (Gänzle & Zheng, 2019; Landis *et al*., 2021).

Back-slopping imposes repeated ecological selection. Early communities are influenced by flour-associated organisms, whereas progressive acidification and substrate depletion favour organisms adapted to low pH, maltose-rich conditions and recurrent dilution. Defined starters improve reproducibility, but spontaneous systems may contain broader metabolic capacity. The relevant design question is therefore not whether spontaneous or defined fermentation is universally superior, but whether a consortium reliably expresses the pathway needed for a specified metabolite in a specified flour matrix.

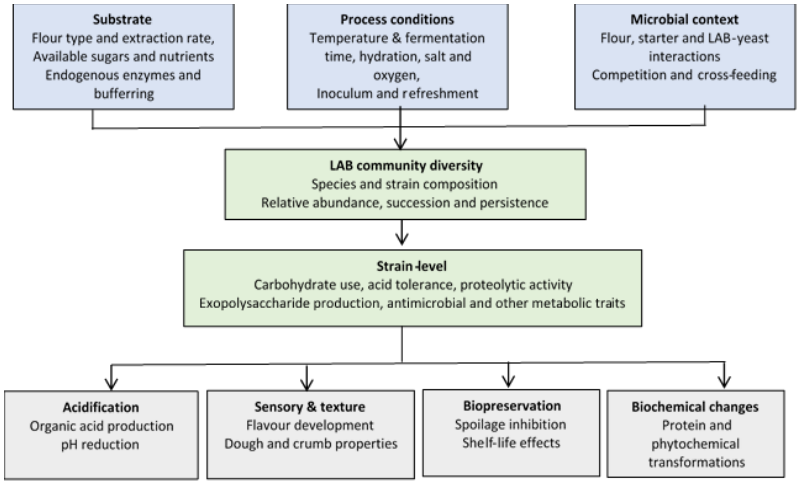


Figure 1: Ecological Determinants and Functional Consequences of LAB Diversity in Sourdough (Calvert et al., 2021)

Process Determinants

Flour establishes the precursor landscape. Whole-grain and bran-rich flours generally supply more phenolic acids, minerals, fibre and endogenous enzymes than refined flour, but higher precursor abundance does not guarantee higher bioavailability. Cell-wall binding, genotype, milling and heat treatment modify accessibility. Non-wheat cereals and pseudocereals expand chemical diversity, but comparisons must distinguish the intrinsic composition of the flour from the effect of fermentation.

Temperature, time, pH and hydration are interdependent rather than isolated variables. Longer fermentation can increase proteolysis, phytate degradation and acid accumulation, but excessive acidification can impair structure or suppress particular enzymes. Higher dough yield increases diffusion and often accelerates microbial metabolism; firmer doughs select for different stress responses and alter the lactic-to-acetic acid ratio. Salt, oxygen and inoculum history further shape the community. Single-factor optimization therefore transfers poorly across cereal matrices.

Starter selection should be based on validated phenotype. A strain reported to produce GABA in broth may fail in dough if free glutamate is limiting; an EPS producer may require sucrose; and a proteolytic strain may generate peptides that disappear during baking or digestion. Factorial designs and response-surface methods are more informative than one-factor-at-a-time experiments because they detect interactions and define an operational window that balances bioactive yield, loaf quality and sensory acceptability.

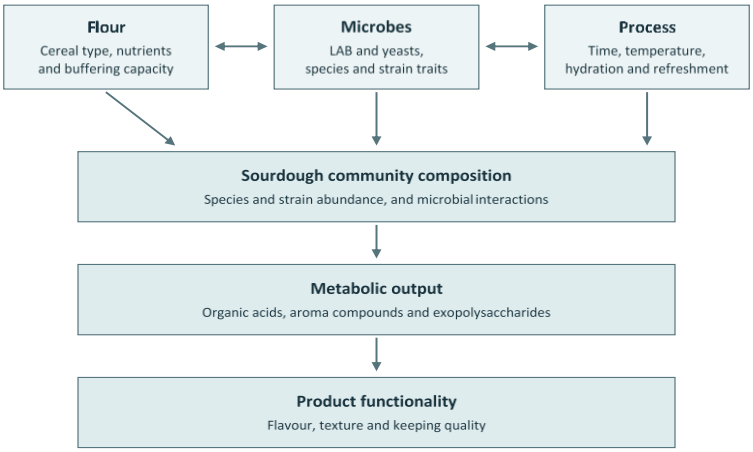


Figure 2: Interacting Determinants of Sourdough Community Composition and Metabolic Output (De Vuyst et al., 2014)

Biosynthesis and Release of Major Bioactive Classes

Across bioactive classes, fermentation changes concentration through four non-equivalent routes: de novo microbial synthesis, enzymatic release from the cereal matrix, conversion of a precursor into a derivative, and apparent enrichment caused by dry-matter loss. These routes should be distinguished using substrate-product mass balances and, where feasible, isotope tracing. Studies of phenolic conversion, protein hydrolysis, GABA formation and antioxidant activity demonstrate both strong strain dependence and substantial effects of cereal substrate and process conditions (Gänzle, 2014; Palla *et al*., 2020; Koistinen *et al*., 2018; Li *et al*., 2024).

The biological meaning of enrichment depends on chemical identity and dose. Total-phenolic assays, radical-scavenging tests and ACE-inhibition screens are useful for discovery but cannot substitute for compound-resolved LC-MS/MS, peptide sequencing, stability testing and exposure measurements. The strongest claims arise when a named molecule is quantified in flour, fermented dough, baked bread and digestate and is linked to activity at a physiologically achievable concentration (Gobbetti *et al*., 2020; Chen *et al*., 2021; Adebo *et al*., 2021; Seyoum *et al*., 2016; Palla *et al*., 2019).

EPS, phytate and mineral outcomes illustrate the importance of separating technological from physiological functionality. In situ dextran or β-glucan can improve rheology and texture, whereas phytate hydrolysis can increase mineral solubility; neither observation alone proves altered absorption or a clinical benefit. Polymer structure, yield per serving and colonic fermentability, as well as mineral speciation and human uptake, remain necessary translational measurements (Dan *et al*., 2025; Brand *et al*., 2025a; Koj & Pejcz, 2023; Montemurro *et al*., 2023).

Organic Acids and Related Carbohydrate Metabolites

Homofermentative LAB converts hexoses mainly to lactate through glycolysis, whereas obligately or facultatively heterofermentative LAB use the phosphoketolase pathway to produce lactate together with acetate or ethanol and carbon dioxide. Electron acceptors, oxygen and co-substrates affect the acetate-to-ethanol balance. Organic acids are technologically important because they lower pH, inhibit spoilage organisms, alter protein and starch interactions and activate cereal enzymes (Perri *et al*., 2021). Their designation as bioactives should nevertheless remain product- and outcome-specific.

Lactic acid can modify starch protein interactions and may slow starch hydrolysis, while acetic acid may influence gastric emptying and postprandial metabolism. These effects cannot be separated easily from flour particle size, fibre, loaf structure and resistant starch. Therefore, studies comparing sourdough with yeast bread should match ingredients, baking loss and available carbohydrate, and should quantify total titratable acidity and individual acids in the consumed portion. Antifungal effects are better supported than systemic health effects and can extend shelf life by combining acidification with microbially generated antifungal metabolites (Quattrini *et al*., 2019).

Phenolic Compounds

Most cereal phenolics are not synthesized de novo by sourdough organisms. Rather, fermentation increases extractability or converts existing compounds. Acidification and cell-wall modification release phenolic acids bound to arabinoxylans, while microbial esterases, glycosidases, reductases and decarboxylases can generate derivatives with different solubility and activity. Ferulic acid is particularly important in wheat and rye, but the direction and magnitude of change depend on genotype, milling and analytical fractionation (free, conjugated and bound).

Higher extractable phenolics frequently increase DPPH, ABTS or FRAP values, yet these assays measure chemical reactions under artificial conditions. They neither reproduce digestion nor establish tissue exposure. Stronger studies quantify identified metabolites before and after fermentation, after baking and following simulated digestion, then relate the resulting concentrations to cellular or human endpoints. Evidence for enhanced chemical antioxidant capacity is strong; evidence that sourdough phenolics improve clinical antioxidant status is limited (Zhao *et al*., 2021).

Bioactive Peptides and Amino-Acid Derivatives

The Dual-Phase Proteolytic Cascade

The generation of bioactive peptides during sourdough fermentation is not a single-enzyme event. Rather, it results from coordinated proteolysis involving endogenous cereal proteases and the proteolytic machinery of sourdough lactic acid bacteria (LAB). Acidification by LAB activates cereal proteases, promotes gluten depolymerization and increases the availability of peptides that can subsequently be metabolized by microbial peptidases (Thiele *et al*., 2004;

Nakamura *et al*., 2007; Gobbetti *et al*., 2020). Acidification and endogenous cereal proteolysis disrupt the macromolecular gluten network, particularly the polymeric glutenin fraction and associated gliadin proteins (Thiele *et al*., 2004). This initial hydrolysis converts parts of the insoluble and highly cross-linked gluten structure into smaller, water- or salt-soluble protein fragments and intermediate peptides (Thiele *et al*., 2004). After cereal proteins have undergone primary hydrolysis, LAB-associated proteolytic enzymes may further degrade the resulting fragments. Depending on the strain, cell-envelope-associated proteinases and extracellular or cell-associated peptidases can hydrolyze accessible proteins and intermediate peptides into smaller oligopeptides (Coda *et al*., 2012; Gobbetti *et al*., 2020).

The resulting peptides can be transported into bacterial cells through oligopeptide, dipeptide and other peptide-transport systems. Once inside the cytoplasm, a complementary array of intracellular peptidases continues peptide hydrolysis:

PepN and PepC, which are general aminopeptidases, progressively release amino acids from the amino terminus of peptide substrates. PepX, an X-prolyl dipeptidyl peptidase, contributes to the degradation of peptides containing proline residues, which are abundant in cereal prolamins. PepT and PepV, which exhibit tripeptidase and dipeptidase activities, respectively, hydrolyze smaller peptide fragments and contribute to the formation of free amino acids.

Structural Determinants of Bioactivity

The biological properties of sourdough-derived peptides depend on their amino-acid sequence, chain length, molecular mass, hydrophobicity and spatial conformation. Chromatographic and mass-spectrometric investigations have identified low-molecular-weight peptides that are generated or enriched during sourdough fermentation (Nakamura *et al*., 2007; Coda *et al*., 2012).

Nevertheless, bioactive sourdough peptides should not be described exclusively as sequences containing 3–10 amino acids. For example, Coda *et al*. (2012) identified antioxidant peptides containing approximately 8–57 amino-acid residues. Therefore, the active molecular-weight and chain-length range varies with the biological activity being investigated.

The ACE-inhibitory potential of food-derived peptides is strongly influenced by their terminal amino-acid composition. Short peptides containing hydrophobic, aromatic or proline residues near the C-terminal region may interact favourably with the ACE active site. Although hydrophobic, aromatic and proline residues near the C terminus frequently strengthen ACE-inhibitory activity in food-derived peptides, a universal sequence rule has not been established for sourdough-derived peptides. Structure-activity relationships should therefore be reported as peptide-specific hypotheses supported by sequence identification, inhibition kinetics and testing after digestion, rather than as fixed compositional requirements.

Selected LAB can release cereal-derived peptides with radical-scavenging and lipid-oxidation-inhibitory activities during sourdough fermentation. The antioxidant capacity of these peptides is associated with their sequence and the presence of amino acids capable of donating electrons or hydrogen atoms, chelating transition metals or stabilizing reactive intermediates (Coda *et al*., 2012). Coda *et al.* (2012) further demonstrated that selected peptide fractions retained antioxidant activity after simulated digestion and protected cultured mouse fibroblasts against experimentally induced oxidative stress. These findings establish peptide formation and in vitro or ex vivo activity but do not, by themselves, demonstrate systemic antioxidant or antihypertensive efficacy in humans.

The Glutamate Decarboxylase Pathway for GABA Enrichment

The biosynthesis of γ-aminobutyric acid (GABA) by certain sourdough LAB is closely associated with their response to environmental acid stress. In GAD-positive strains, the glutamate decarboxylase system converts L-glutamate into GABA while consuming an intracellular proton, thereby contributing to cytoplasmic pH regulation and microbial persistence under acidic sourdough conditions (Su *et al*., 2011).

The system operates through a coupled decarboxylation and antiport mechanism:

i. The membrane-associated glutamate/GABA antiporter, GadC, transports extracellular glutamate into the bacterial cytoplasm.
ii. Intracellular glutamate decarboxylase catalyses the pyridoxal 5′-phosphate-dependent decarboxylation of glutamate. This reaction consumes a cytoplasmic proton and generates GABA and carbon dioxide.
iii. The resulting GABA is exported through GadC in exchange for another extracellular glutamate molecule.

Through proton consumption and product exchange, the GAD system helps LAB maintain intracellular pH homeostasis and improves survival and competitiveness during acidic sourdough fermentation. Experimental evidence from *Lactobacillus reuteri* currently classified as *Limosilactobacillus reuteri* demonstrates that glutamate-dependent GABA formation contributes to acid resistance and persistence in industrial sourdough ecosystems (Su *et al*., 2011).

The amount of GABA accumulated in sourdough depends on the LAB strain, availability of glutamate or glutamine, fermentation pH, temperature and duration. Therefore, substantial differences can occur among cereal substrates and fermentation protocols (Su *et al*., 2011; Gobbetti *et al*., 2020).

Translational Evidence Barriers for Peptides and GABA

Although sourdough fermentation can release peptides with measurable biochemical activity, formation in fermented dough represents only an early level of functional evidence. Demonstrating physiological efficacy additionally requires evidence of processing stability, gastrointestinal survival, intestinal absorption, target-tissue exposure and reproducible human outcomes.

Baking may modify the amount, structure and accessibility of fermentation-derived peptides. Heat exposure can promote additional proteolysis, peptide degradation, aggregation and reactions between amino groups and reducing sugars. These transformations may reduce the recoverability of some peptides, although their effects depend on peptide sequence, location within the bread and baking conditions. More broadly, sourdough-mediated biochemical changes contribute to bread structure and technological properties, which may influence the physical accessibility of protein-derived compounds (Arendt *et al*., 2007; Gobbetti *et al*., 2020).

Peptides that remain in baked bread are subsequently exposed to gastric, pancreatic and intestinal brush-border enzymes. Digestion may destroy an originally active sequence, preserve it or release new bioactive fragments from larger precursor peptides. Importantly, Coda *et al*. (2012) reported that selected sourdough-derived antioxidant peptide fractions retained activity following in vitro digestive-enzyme treatment. Thus, gastrointestinal degradation should not be presented as universal; peptide stability must be experimentally determined for each identified sequence.

Comparable caution is required when interpreting GABA enrichment. GAD-positive LAB can produce GABA and gain

a competitive advantage under acidic sourdough conditions, but the final GABA concentration depends strongly on strain selection and precursor availability (Su *et al*., 2011). Evidence of GABA formation in fermented dough does not establish that a baked serving delivers a clinically effective dose.

Exopolysaccharides

Sourdough LAB form homopolysaccharides such as dextran and levan from sucrose through extracellular glucansucrases or fructansucrases, and heteropolysaccharides through intracellular sugar-nucleotide pathways and membrane-associated polymerization. Polymer yield and branching vary by strain, sucrose supply, temperature and pH. This structural diversity is central: molecular weight and linkage pattern influence water binding, viscosity, fermentability and biological activity (Caggianiello *et al*., 2016; Lynch *et al*., 2018a, 2018b; Schmid *et al*., 2018).

The most defensible benefits of in situ EPS production are technological. EPS can improve gas retention, crumb softness and shelf life and can partly replace hydrocolloids in gluten-free bread. Prebiotic, immunomodulatory and antioxidant effects are biologically plausible but are often derived from purified polymers, non-sourdough strains or high experimental doses. A claim about the final bread requires polymer quantification after baking, structural characterization and evidence of selective microbial utilization or a host response at the consumed dose.

Vitamins, Phytate Reduction and Antimicrobial Metabolites

Selected LAB and yeasts can synthesize folate, riboflavin and other B vitamins, but enrichment is strain- and medium-dependent and may be offset by microbial consumption or heat loss. Vitamin B12 claims require exceptional caution because many plant-fermentation studies measure corrinoid analogues rather than bioactive cobalamin. Validated chromatographic or microbiological methods and baking-retention data are required. Phytate reduction is among the most reproducible nutritional consequences of sourdough acidification. A lower pH activates cereal phytases and some microorganisms contribute phytase activity, releasing phosphate and reducing mineral chelation. Greater soluble or dialysable mineral content indicates improved bioaccessibility, but mineral status depends on the total diet and should not be inferred from an in vitro assay alone.

Organic acids, cyclic peptides and other low-molecular-weight compounds can suppress moulds and rope-forming bacteria. These activities support clean-label preservation, although efficacy depends on concentration, dissociation at product pH, water activity and microbial target. Bacteriocin activity demonstrated in culture does not necessarily persist in dough because proteases, adsorption and heat can reduce activity.

Table 2: Biochemical Origins, Reported Effects and Translational Limitations of Sourdough Bioactives

Bioactive Class	Origin/Pathway	Outcome	Major Translational Uncertainty	References
Organic acids	Hetero- and homofermentative carbohydrate metabolism yielding variable lactic-to-acetic acid ratios via phosphoketolase or glycolytic paths.	Systematically depresses dough pH, which activates native cereal proteases. Delays retrogradation and retards fungal spoilage.	Volatilization of acetic acid during high-temperature baking. Dispersal effects are difficult to separate from the macrostructural changes of the crumb.	(Gänzle, 2014; Palla *et al*., 2020; Thiele *et al*., 2004)
Phenolic derivatives	Hydrolysis of ester-linked phenolics bound to cell-wall arabinoxylans via microbial ferulic acid esterases. Reduction or decarboxylation to highly active volatile derivatives.	Shifts insoluble-bound grain phenolics into highly extractable free fractions. Augments baseline *in vitro* radical scavenging values.	Rapid chemical degradation or irreversible polymerization via Maillard cross-linking in the bread crust. Rapid hepatic conjugation limits free systemic tissue exposure.	(Liukkonen *et al*., 2003; Koistinen *et al*., 2018; Li *et al*., 2024)
Peptides	Primary breakdown of macro-gluten strings by native cereal aspartic peptidases, followed by cell-envelope serine proteinases (CEPs).	Release of peptides with sequence-dependent in vitro activity; antioxidant peptides of 8-57 amino-acids residues were identified.	Extensive cytoplasmic over-hydrolysis by redundant LAB aminopeptidases into basic free amino acids. Luminal and brush-border degradation during digestion.	(Coda et al., 2012; Arendt *et al*., 2007; Gobbetti *et al*., 2020; Chen *et al*., 2021)
GABA	Electrogenic antiport decarboxylation of L-glutamate driven by the homohexameric enzyme glutamate decarboxylase (GAD) to survive acid shock.	Intracellular proton consumption step that enriches the dough with γ-aminobutyric acid, providing a putative neuro-metabolic signal.	Precursor availability, strain-dependent production and retention after baking; the retained dose per serving and its clinical relevance require product-specific verification	(Adebo *et al*., 2021; Seyoum *et al*., 2016; Palla *et al*., 2019)
EPS	Cell-surface glucansucrase or fructansucrase mediated synthesis using sucrose to construct high-molecular-weight homopolymers (dextrans/levans).	Improved crumb softness and storage quality in selected formulations; potential hydrocolloid replacement requires	Structural branching configurations alter colonic fermentability. Claims regarding host immunomodulation are often extrapolated from	(Dan *et al*., 2025; Brand *et al*., 2025a; Koj & Pejcz, 2023; Galle *et al*., 2012)

Bioactive Class	Origin/Pathway	Outcome	Major Translational Uncertainty	References
		formulation-specific validation.	irrelevant cell-culture contexts.	
Vitamins	Strain-specific microbial synthesis	Possible enrichment of selected B vitamins	Analytical specificity and thermal retention	(Lopez *et al*., 2001; Perri *et al*., 2021; Montemurro *et al*., 2023)
Phytate reduction	Cereal and microbial phytases promoted by acidification	Improved mineral bioaccessibility	Long-term mineral status in humans	(Abedfar *et al*., 2020; Nieto *et al*., 2024)

Formation during fermentation does not establish retention or biological activity after baking. Retention must be measured for the specific compound, formulation and processing conditions. For example, Verni et al. (2022) reported approximately 10% GABA loss during baking in their experimental bread; this finding should not generalized to other compounds or breads.

Processing Stability and Gastrointestinal Fate

Baking is a decisive intervention rather than a neutral finishing step. Thermal gradients differ between crumb and crust, moisture is redistributed, microbial metabolism stops, enzymes are inactivated and Maillard chemistry creates and removes measurable compounds. Studies that analyse only sourdough or proofed dough can therefore establish biosynthesis but not consumer exposure. Bread-level studies show that fermentation time, leavening system and baking conditions jointly influence digestibility, phenolic recovery, acrylamide formation and nutritional quality (Won *et al*., 2022; Carnevali *et al*., 2007; Liszkowska *et al*., 2024).

Post-baking storage should also be incorporated because starch retrogradation, moisture migration and continued oxidation can change glycaemic properties and compound recovery. A minimum mass-balance design should report values on both wet- and dry-weight bases, loaf yield, serving size, analytical recovery and the percentage retained after baking and storage (Munch-Andersen *et al*., 2025; Ameur *et al*., 2024; Nasiri Esfahani *et al*., 2017).

During digestion, release from the matrix may increase even when the parent compound decreases, because peptides and phenolics are transformed into absorbable or colon-available products. Static digestion is suitable for screening, but dynamic digestion, epithelial transport, colonic fermentation and targeted human metabolomics answer different questions and should not be presented as interchangeable evidence (Petrova & Petrov, 2020; Carbonetto *et al*., 2018; Montemurro *et al*., 2020; Fu *et al*., 2024; Rinaldi *et al*., 2017; Montemurro *et al*., 2019; Kim *et al*., 2025).

Baking Changes the Exposure

Fermentation studies often stop at proofed dough, whereas consumers ingest baked and stored bread. Crumb temperatures approach the boiling point of water and the crust experiences substantially higher temperatures. Heat kills most starter organisms, inactivates enzymes, denatures proteins and accelerates Maillard chemistry. Organic acids partly volatilize; peptides may be hydrolysed, cross-linked or newly released; and vitamin retention differs by compound. Phenolic extractability can rise through matrix disruption even as individual molecules degrade (Gobbetti *et al*., 2014).

The final concentration should therefore be reported per gram of bread and per realistic serving, not only per gram of dough or as a percentage increase. Moisture loss can create an apparent concentration increase. Appropriate mass-balance reporting measures the precursor in flour, fermented dough and baked bread, accounts for water loss, and distinguishes absolute retention from extractability. Storage should also be considered because moisture redistribution, starch retrogradation and continued chemical reactions alter digestibility and texture.

Digestion, Absorption and Colonic Metabolism

Oral, gastric and intestinal digestion changes the food matrix sequentially. Acid conditions and pepsin extend protein hydrolysis; pancreatic enzymes release peptides and amino acids; bile and intestinal conditions alter the solubility of phenolics; and non-digestible carbohydrates pass to the colon. Static digestion models are useful for screening but do not capture transit, absorption or host variation. Dynamic models and intestinal transport systems can strengthen inference when linked to targeted chemical analysis.

Bioaccessibility is the fraction released from the matrix and available for intestinal uptake; bioavailability additionally requires absorption, distribution, metabolism and measurable exposure. These terms should not be used interchangeably. Phenolics may be conjugated rapidly in the intestinal wall and liver, while EPS and fibre-associated compounds are more likely to act through colonic fermentation. Microbial conversion in the colon can yield smaller phenolic metabolites or short-chain fatty acids, but attributing those metabolites specifically to sourdough requires matched bread controls.

A rigorous pipeline would combine compositional analysis, standardized digestion, intestinal transport or fermentation models, and plasma, urine or faecal metabolomics in human studies. The analysis should test whether fermentation creates a unique exposure rather than merely increasing an extractable signal in vitro.

Mechanisms of Action: Evidence and Boundaries

Mechanistic attribution requires triangulation. A pathway-level response in cells should be reproduced using digested bread at a realistic dilution, linked to a quantified candidate metabolite, blocked or attenuated by an appropriate inhibitor, and then associated with exposure in vivo. Without this sequence, changes in antioxidant, inflammatory or vascular markers remain compatible with multiple properties of the food matrix (Gänzle, 2014; Palla *et al*., 2020; Liukkonen *et al*., 2003; Koistinen *et al*., 2018; Li *et al*., 2024; Arendt *et al*., 2007; Chen *et al*., 2021; Adebo *et al*., 2021).

For microbiota-mediated mechanisms, the relevant exposure is usually the fraction that survives small-intestinal digestion and reaches the colon. Changes in fibre accessibility, resistant starch, EPS and residual fermentable carbohydrates can modify microbial metabolism, but conventional baked sourdough bread should not be described as probiotic. Any postbiotic claim should satisfy the accepted requirement for a preparation of inanimate microorganisms and/or their components that confers a demonstrated health benefit.

Antioxidant and Inflammatory Pathways

Phenolic metabolites and some peptides can scavenge radicals, chelate transition metals or influence endogenous antioxidant responses. Cell studies suggest potential modulation of Nrf2-regulated defences and inflammatory pathways such as NF-κB, but direct evidence using digested sourdough at physiologically plausible concentrations is limited. Claims that sourdough activates a named signalling pathway should therefore identify the active molecule, exposure concentration and experimental model.

Inflammation may also be influenced indirectly through glycaemic exposure, intestinal barrier function or microbial fermentation of fibre. Such effects arise from the whole food matrix and cannot automatically be attributed to a specific fermentation-derived metabolite. Chemical antioxidant assays should be presented as compositional descriptors, not as evidence of disease prevention.

Vascular and Metabolic Mechanisms

ACE-inhibitory peptides and GABA provide plausible routes to vascular effects, while organic acids and altered bread structure may modify postprandial glycaemia. Nevertheless, evidence is strongest for acute metabolic responses and weakest for cardiovascular events. Blood-pressure or endothelial claims require adequate dosing, sequence-defined peptides or a characterized food exposure, and controlled human measurement.

The glycaemic response reflects several interacting variables: available carbohydrate, flour particle size, fibre, organic acids, starch gelatinization and retrogradation, loaf density, chewing and host physiology. Fermentation is one contributor rather than a sufficient explanation. Matched comparators are essential; comparing whole-grain sourdough with industrial white bread confounds fermentation with grain composition and processing.

Gut Microbiota and Immune Mechanisms

Most sourdough organisms are not expected to remain viable after baking, so probiotic terminology is generally inappropriate for conventional bread. Fermentation-derived metabolites, inactivated cells and cell components may resemble postbiotic preparations, but the consensus definition of a postbiotic requires a preparation of inanimate microorganisms and/or their components that confers a health benefit; the label should not be applied merely because microbes were once present (Salminen *et al*., 2021).

Sourdough can alter fermentable carbohydrates, arabinoxylan accessibility, resistant starch and EPS, which may influence colonic microbiota. However, broader fermented-food trials cannot establish a sourdough-specific effect. The influential fermented-food intervention by Wastyk *et al*. (2021), for example, investigated a diet rich in several fermented foods rather than sourdough alone. Such work supports plausibility but should not be cited as direct evidence for sourdough bread.

Human Health Evidence

Table 3 summarizes representative human interventions and deliberately retains null findings. Overall certainty is low for a sourdough-specific improvement in glycaemia, satiety or gastrointestinal symptoms because most trials are small, acute and incompletely matched for grain type, fibre, loaf structure or acidity. Certainty is very low for sustained cardiovascular, immune or microbiome benefits. The evidence supports product-specific effects, not a class-wide health claim.

Table 3: Representative Human Intervention Studies of Sourdough Products, Including Null Findings and Design Limitations

Study/Design	Population	Sourdough Exposure	Comparator	Principal Outcome and Result	Appraisal	References
Acute randomized crossover	Healthy adults (n=15)	Mixed wheat sourdough bread with oat fibre	White-wheat bread/glucose reference	GI 53.7 versus 72; formulation combined acidity and fibre	Small sample; ingredient and fermentation effects not separable	(De Angelis *et al*., 2007)
GI evaluation	Healthy adults; sample not stated in abstract	Two commercial sourdough/intact-grain prototypes	Glucose reference	In-vivo GI 65 and 67; both medium-GI with low glycaemic load	Prototype-specific; no yeast-bread comparator	(Hefni & Witthöft, 2021)
Randomized crossover breakfast	Healthy adults (n=24)	Sourdough whole-grain rye crispbread	Unfermented rye and yeast-refined-wheat crispbreads	Lower hunger than refined wheat, but insulin response lowest after unfermented rye	Matrix differed by rye/refinement; sourdough not independently isolated	(Zamaratskaia *et al*., 2017)
Randomized crossover breakfast	Healthy volunteers (n=23)	Five breads containing 9–51% sourdough and 35–48% rye	Yeast-fermented refined-wheat bread	No consistent effect on appetite or ad-libitum intake	Multiple formulation differences; acute endpoint	(Iversen *et al*., 2018)
In-vivo postprandial challenge	Healthy volunteers (n=36)	Two sourdough breads differing in fermentation intensity	Baker's-yeast bread	Faster gastric emptying and lower glycaemia; satiety differed between sourdoughs	Acute, product-specific, multiple physicochemical differences	(Rizzello *et al*., 2019)

Study/Design	Population	Sourdough Exposure	Comparator	Principal Outcome and Result	Appraisal	References
Double-blind crossover pilot	Coeliac disease (n=17)	Long-fermented wheat/non-toxic-flour bread; ~2 g gluten	Baker's-yeast bread	Intestinal-permeability response attenuated in 13 responders	Very small acute challenge; not evidence of routine coeliac safety	(Di Cagno *et al*., 2004)
Double-blind controlled study	IBS/wheat sensitivity (n=26)	Long-fermented sourdough wheat bread, lower in FODMAPs/ATIs	Yeast-fermented wheat bread	No improvement in GI symptoms or inflammation markers	Short 7-day study; compositional reduction did not translate clinically	(Laatikainen *et al*., 2017)
Randomized double-blind crossover	IBS; sample not reported in abstract	Reduced-gluten bread/pasta produced with sourdough LAB and fungal proteases	Traditional bread/pasta	VAS improved, but IBS severity, anxiety/depression and quality of life did not	Composite bread-and-pasta intervention; small pilot and short periods	(Calasso *et al*., 2018)
Repeated-meal comparison	GDM (n=43) and healthy pregnancy (n=38)	Whole-grain sourdough bread	Refined white bread of equal weight	Lower 1-h glucose, insulin and C-peptide; 2-h glucose similar	Whole grain/refinement confounding; consecutive-week design	(Özer *et al*., 2023)
Double-blind parallel trial	Metabolic syndrome (n=31)	48-h versus 2-h sourdough bread for 2 months	Short versus long fermentation	No microbiota difference; longer fermentation gave no added benefit	Small sample; no non-sourdough control	(Pérez-Vega *et al*., 2024)
Randomized controlled parallel trial	Healthy adults (n=32)	Fibre-enriched sourdough croissant for 2 weeks	Conventional sourdough croissant	Small improvement in fasting glucose; no gut-microbiome change	Fibre blend is the main exposure contrast; short study	(Barone Lumaga *et al*., 2024)
Two randomized double-blind crossover studies	Self-reported NCWS (n=20 per fermentation study)	Sourdough breads from bread wheat, spelt or emmer	Within-fermentation wheat-species comparisons	Symptoms varied, without a uniform sourdough advantage	Parallel fermentation studies limit direct yeast-versus-sourdough inference	(de Graaf *et al*., 2024)
Double-blind randomized crossover	Healthy adults (n=44)	Wholemeal sourdough, yeast, or combined-leavening bread; 2 weeks each	Matched wholemeal leavening conditions	No difference in glucose, energy intake, gastric emptying or microbiota; minor hormone differences	Strong crossover design; findings argue against a general sourdough effect	(Chatonidi *et al*., 2026)

Clinical evidence remains product-specific. Acute glycaemic studies differ in flour refinement, bread structure, acidity, available carbohydrate and comparator, while gastrointestinal studies differ in participant phenotype, symptom definition and FODMAP or gluten characterization. These differences explain why favourable results cannot be generalized to all sourdough breads (Gobbetti *et al*., 2016; Ravisankar *et al*., 2021; Kwak *et al*., 2026; Gobbetti *et al*., 2019; Gabriele *et al*., 2019; von Gastrow *et al*., 2023; Verni *et al*., 2024; Cevallos-Fernández *et al*., 2026; Di Nunzio *et al*., 2018; Taheri *et al*., 2026; Koistinen *et al*., 2022; Svensson *et al*., 2010; Terrazas-Avila *et al*., 2024; de Graaf *et al*., 2024).

The next generation of trials should pre-register a primary endpoint, chemically characterize the final bread, match available carbohydrate and physical structure, measure adherence and background diet, and use adequate washout in crossover designs. Longer studies are needed to distinguish an acute postprandial effect from durable changes in glycaemic control, blood pressure, gastrointestinal symptoms or microbiome function (Costantini *et al*., 2023; Rizzello *et al*., 2015; Danesi *et al*., 2020; Canlı *et al*., 2021; Liljeberg *et al*., 1995; Reese *et al*., 2020; Novotni *et al*., 2011; Chen *et al*., 2023; Gil-Cardoso *et al*., 2021; Smith *et al*., 2020).

Glycaemic Regulation

Postprandial glycaemia is the most studied clinical endpoint. Some acute trials report lower glucose or insulin responses after sourdough bread, especially when acidity, whole grains or specific processing conditions are used; others show no advantage. The systematic review by Ribet *et al*. (2023)

concluded that heterogeneity prevented a general sourdough effect. Korem *et al*. (2017) found person-specific responses to whole-grain sourdough and white bread, emphasizing that host and product characteristics can dominate the average comparison.

Evidence from reformulated breads more broadly should be interpreted separately. A meta-analysis of 22 trials involving 1,037 participants reported a small reduction in fasting glucose but no consistent effect on insulin, HOMA-IR, HbA1c or postprandial glucose, and the interventions included fibre, whole grains and functional ingredients rather than sourdough alone (Schadow *et al*., 2023). Thus, the most defensible conclusion is that carefully formulated sourdough products can modify acute glycaemia, but fermentation is not universally sufficient and long-term glycaemic benefit remains uncertain.

Gastrointestinal Tolerance and Digestibility

Acidification and microbial enzymes can reduce selected fermentable carbohydrates and partially hydrolyse gluten proteins. This may improve tolerance in some individuals with functional gastrointestinal symptoms. In vivo work has reported different gastrointestinal responses to sourdough and baker's-yeast breads, but small samples and product-specific formulations limit generalization (Rizzello *et al*., 2019). Conventional sourdough bread containing gluten is not safe for people with coeliac disease unless it independently meets the regulatory threshold for gluten-free food.

Claims about improved digestibility should specify the endpoint: gastric emptying, symptom score, protein digestibility, FODMAP exposure and coeliac safety are different questions. Long fermentation alone does not guarantee a low-FODMAP or gluten-free product; strain capability, substrate and analytical verification are required.

Cardiovascular, Mineral and Immune Outcomes

Direct clinical evidence for cardiovascular protection is sparse. Acute glycaemia, in vitro ACE inhibition and chemical antioxidant activity are surrogate observations, not evidence of reduced hypertension, myocardial infarction or mortality. Current data do not justify disease-prevention claims. Mineral bioaccessibility may improve through phytate degradation, but long-term trials measuring mineral status are limited. Evidence for immune modulation and microbiome change is even less direct. Studies of mixed fermented-food diets and purified EPS provide hypotheses but cannot quantify the effect of sourdough bread. Future trials should use pre-specified outcomes, adequate washout in crossover designs, dietary control, blinded outcome assessment where possible and full chemical characterization of both intervention and comparator breads.

Table 4. Endpoint-specific Evidence, Certainty Rationale and Permissible Interpretation of Sourdough-related Effects

Outcome	Endpoint-specific evidence	Certainty and rationale	What can presently be claimed	References
Acute glycaemia	Human feeding studies report product-specific responses. A randomized crossover comparison of wholemeal breads found no difference in glucose responses between leavening conditions.	Low for a general sourdough-specific benefit. Findings vary across formulations, comparators and processing conditions; short interventions limit inference about sustained glycaemic control.	Some sourdough formulations may modify postprandial glucose responses, but general superiority over comparable yeast-leavened bread is not established.	Rizzello et al. (2019); Chatonidi et al. (2026).
Gastrointestinal comfort	A small controlled trial of sourdough wheat bread found no improvement in gastrointestinal symptoms. A separate intervention combined reduced-gluten bread and pasta, providing indirect evidence for sourdough bread alone.	Low for symptom improvement. Small samples, short interventions and differences in food composition limit precision and attribution to sourdough fermentation.	Changes in fermentable carbohydrates or protein composition do not necessarily improve symptoms. Any tolerance benefit must be demonstrated for the specific product and population.	Laatikainen et al. (2017); Calasso et al. (2018).
Mineral bioaccessibility	Experimental sourdough processing reduced phytate and increased magnesium and phosphorus solubility. These measurements concern the food matrix rather than long-term human mineral status.	Mechanistic support; clinical certainty not established by the cited study. Mineral solubility does not directly demonstrate intestinal absorption or improved nutritional status.	Sourdough fermentation can reduce phytate and increase mineral solubility under specified processing conditions. Improved long-term mineral status cannot be inferred from these measurements alone.	Lopez et al. (2001).
Blood pressure and cardiovascular outcomes	An isolated sourdough-derived peptide demonstrated in vitro angiotensin-converting enzyme inhibition. This is mechanistic evidence rather than a clinical bread intervention.	Very low for clinical benefit from this evidence. An enzyme assay does not establish peptide retention, absorption, effective exposure or blood-pressure reduction in humans.	Specific sourdough-derived peptides can exhibit in vitro enzyme-inhibitory activity; this does not establish antihypertensive efficacy or cardiovascular disease prevention after bread consumption.	Nakamura et al. (2007).

Outcome	Endpoint-specific evidence	Certainty and rationale	What can presently be claimed	References
Antioxidant and anti-inflammatory effects	Sourdough-derived peptide fractions showed antioxidant activity in chemical and cellular models. Human bread interventions have also measured inflammatory outcomes, which must be interpreted separately from antioxidant assays.	Mechanistic support for antioxidant activity; low certainty for a general human anti-inflammatory benefit. Experimental models are indirect, while clinical studies are small and compare specific products or fermentation conditions.	Selected sourdough-derived fractions show experimental antioxidant activity. Systemic antioxidant or anti-inflammatory benefits require direct confirmation in human interventions.	Coda et al. (2012); Laatikainen et al. (2017); Pérez-Vega et al. (2024).
Gut microbiota and immune function	Trials comparing fermentation durations or wholemeal bread leavening conditions did not demonstrate a consistent microbiota advantage. These comparisons do not directly establish improved immune function.	Low for a general microbiota benefit; immune benefit not established by these trials. Limited sample sizes, intervention durations and comparator scope restrict generalization.	A reproducible sourdough-specific improvement in gut microbiota composition is not established. Microbiota findings alone do not demonstrate improved immune function.	Pérez-Vega et al. (2024); Chatonidi et al. (2026).

Omics and Predictive Fermentation

Causal multi-omics should connect genes to transcripts, enzymes and metabolites across fermentation time. Dense time-series sampling, absolute rather than solely relative abundance, strain-resolved metagenomics and targeted metabolite validation can reveal which community members produce, consume or transform each compound. Metatranscriptomic and synthetic-community studies demonstrate the value of resolving active pathways rather than inferring function from taxonomy alone (Calabrese *et al*., 2022; Menezes *et al*., 2020; Boreczek *et al*., 2020; Leroy *et al*., 2006; Nakamura *et al*., 2007; Dentice Maidana *et al*., 2020; Arora *et al*., 2022; Pontonio *et al*., 2016; De Angelis *et al*., 2019).

Amplicon sequencing describes community composition, shotgun metagenomics predicts functional potential, metatranscriptomics measures expressed genes, proteomics characterizes enzymes and peptides, and metabolomics captures chemical output. Their greatest value lies in integration. Taxonomic abundance should be linked to expressed pathway, quantified metabolite and process condition, ideally with isolate genomes and synthetic-community experiments.

Single-omics associations can be misleading because a gene may not be expressed and a detected metabolite may derive from flour chemistry rather than microbial synthesis. Stable-isotope tracing and targeted perturbation provide stronger causal evidence. Standardized metadata flour genotype, extraction rate, dough yield, inoculum, temperature, pH trajectory, time and baking profile—are essential for cross-study reuse.

Machine learning may predict acidification, community succession or metabolite yield, but current sourdough datasets are often too small and heterogeneous for robust generalization. Model performance should be tested on independent batches and external laboratories; leakage between technical replicates must be prevented; and interpretable variables should guide experimental validation. Digital twins remain a promising engineering direction rather than established evidence of health benefit.

Research Priorities and Translational Roadmap

A high-value translational programme would use a staged decision gate: demonstrate reproducible production; verify retention after baking and storage; establish bioaccessibility and exposure; confirm mechanism at an achievable concentration; and only then undertake a powered human trial. Failure at an earlier gate should redirect strain or process optimization rather than be concealed by adding more nonspecific antioxidant assays.

Table 5: Priority Experiments Required to Advance Sourdough Bioactives from Compositional Findings to Validated Health Effects

Priority	Recommended Design	Reason	References
Define the exposure	Report strain, flour, process, baking profile and metabolite dose per serving	Makes studies reproducible and clinically interpretable	(Won *et al*., 2022; Carnevali *et al*., 2007; Liszkowska *et al*., 2024)
Follow mass balance	Measure precursor/metabolite in flour, dough, bread and digesta	Separates formation, concentration and survival	(Munch-Andersen *et al*., 2025; Ameur *et al*., 2024; Nasiri Esfahani *et al*., 2017)
Use matched controls	Match grain, milling, recipe, loaf structure and available carbohydrate	Isolates fermentation from formulation	(Weckx *et al*., 2019; De Vuyst *et al*., 2017; De Vuyst *et al*., 2023)
Identify active entities	Sequence peptides; characterize EPS linkages; quantify phenolic metabolites	Prevents claims based on nonspecific extracts	(Oshiro *et al*., 2021; Yu *et al*., 2021; De Vuyst *et al*., 2016)

Priority	Recommended Design	Reason	References
Test bioavailability	Link digestion to transport, colonic fermentation and human metabolomics	Establishes plausible tissue or microbial exposure	(Clark *et al*., 2024; Minervini *et al*., 2014; De Vuyst *et al*., 2014)
Strengthen trials	Adequately powered randomized designs with preregistered endpoints	Moves from surrogate assays to causal human evidence	(Gänzle *et al*., 2023; Zhour *et al*., 2026; Cocolin *et al*., 2013)
Validate prediction	External validation of multi-omics and machine-learning models	Avoids overfitting and enables industrial transfer	(Meroth *et al*., 2003; Menezes *et al*., 2024; Coda *et al*., 2012)

Industrial development should begin with a target product specification rather than a generic ambition to maximize bioactives. A starter selected for EPS may improve softness but change sweetness or acid perception; extensive proteolysis may increase peptides while weakening dough; and increased acidity may impair consumer acceptance. Multi-objective optimization is therefore required to balance metabolite retention, safety, sensory quality, shelf life and cost.

Regulatory communication should distinguish nutritional composition, technological function and health effect. Terms such as antioxidant, postbiotic, probiotic and low-FODMAP carry different evidentiary or regulatory implications. Until controlled trials support specific endpoints, product communication should emphasize verified composition and process attributes rather than chronic-disease prevention.

The field would benefit from reference sourdough systems and minimum reporting standards. Shared materials, harmonized digestion protocols, authenticated starter collections and open metabolomics data could reveal which findings are general and which are product-specific. The most informative future studies will connect microbial ecology to chemical mass balance and human exposure in the same experimental programme.

CONCLUSION

Sourdough fermentation can transform cereals in nutritionally and technologically meaningful ways. Strong evidence supports acidification, phytate reduction, release or conversion of phenolic compounds and the technological value of selected microbial exopolysaccharides. Production of peptides, GABA and vitamins is feasible but highly dependent on strain, substrate and process. None of these changes should be equated automatically with a physiological benefit. The decisive translational questions occur after fermentation: whether the compound remains in the baked bread, becomes bioaccessible during digestion, produces a biologically relevant exposure and improves a validated human endpoint. Current human evidence is concentrated on acute glycaemia and gastrointestinal responses and is too heterogeneous to establish that sourdough per se prevents cardiovascular, metabolic or inflammatory disease. This review is limited by its structure but non-systematic search, possible publication and language bias arising from reliance on published English language studies, and subjectivity in the narrative grading of evidence.

Accordingly, sourdough should be understood as a controllable biotransformation platform rather than a uniform health category. Progress will depend on chemically characterized final products, matched comparators, strain-resolved microbiology, processing and digestion mass balances, and adequately powered human trials. This evidence-chain approach can guide the testing of product-specific health claims, which require confirmation through adequately powered, well-controlled human trials before they can be considered established

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