Journal of *Phytomedicine*

MDPI

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Microbially Matured Phytomedicines from Sesame Hull (*Sesamum indicum* L.) Cell-Wall Oligosaccharides: *Lactobacillus*-Generated Pre-Postbiotics with Antioxidant, Enzyme-Inhibitory and Anti-*Helicobacter pylori* Activity in a Functional Beverage

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Abstract

Many bioactive constituents of medicinal plants depend on microbial biotransformation for their pharmacological activity, positioning postbiotics from plant substrates as *microbially matured phytomedicines*. An emerging framework integrates prebiotic phytochemicals with probiotic strains to modulate gut microbiota and host health. In this study, we explored the functional properties of heat-inactivated *Lactobacillus* strains following the fermentation of oligosaccharides obtained from sesame hulls (*Sesamum indicum* L.), underutilised agro-industrial residues. Cell-wall oligosaccharides were obtained by alkaline or enzymatic (Celluclast® 1.5 L (Novonesis, Copenhagen, Denmark)) extraction with Ultraflo® L (Novonesis, Copenhagen, Denmark) hydrolysis and fermented with *Lactobacillus acidophilus*, *L. casei*, or *L. paracasei*. Heat-inactivated pre-postbiotic preparations were profiled for antioxidant capacity, inhibition of metabolic enzymes implicated in obesity and type 2 diabetes, and anti-*Helicobacter pylori* urease activity. Moreover, these preparations were incorporated into a barley malt (*Hordeum vulgare* L.) beverage. Bioactivity was strain- and substrate-dependent: *L. casei*-derived postbiotics most strongly inhibited pancreatic lipase (47.82%) and α-glucosidase (52.14%); *L. acidophilus* most strongly inhibited α-amylase (43.67%); and *L. paracasei* exhibited the strongest urease inhibition (20.66%). All strains displayed enhanced antioxidant activity, with ABTS scavenging reaching 87.02%. The supplemented beverages improved antioxidant activity by ~20%. The fermentation of these oligosaccharides thus yields a microbially matured phytomedicine with multi-target activity, supporting postbiotics as active mediators of plant-based therapeutics.

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Academic Editor: Michał Tomczyk

Received: 29 April 2026
Revised: 1 June 2026
Accepted: 5 June 2026
Published: 30 June 2026

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Keywords: phytomedicine; postbiotics; microbial biotransformation; xylo-oligosaccharides; *Lactobacillus*; *Sesamum indicum*; sesame hull valorisation; α-glucosidase inhibition; anti-*Helicobacter pylori*; functional beverage

1. Introduction

The classical conception of phytomedicine, defined by the World Health Organisation as therapy with herbal medicines and their preparations [1], has historically focused on the chemical complement of plant material itself. Yet a substantial body of pharmacological evidence now indicates that many of the bioactive constituents of medicinal plants polyphenols, glycosides, and structural polysaccharides are weakly active or pharmacologically inert in their native form, and that their therapeutic activity depends on microbial biotransformation in the gastrointestinal tract or during fermentation processing [2–4]. Polyphenols are converted by gut microbiota into smaller phenolic acids [2]; glycosides are hydrolysed to aglycones [3]; and structural fibres including (arabino-)xylans are fermented into short-chain fatty acids and other bioactive metabolites [4,5]. The resulting compounds are, by definition, postbiotic products [6], yet they are structurally derived from plant substrates and functionally responsible for the pharmacological effects historically attributed to the parent phytomedicine.

Postbiotics formally defined by the International Scientific Association of Probiotics and Prebiotics (ISAPP) as "*preparations of inanimate microorganisms and their components that confer a health benefit on the host*" [6] comprise inactivated microbial cells, cell wall components, and metabolites [7,8]. They have advantages over live probiotics in stability, safety in vulnerable populations (immunocompromised, post-surgical, premature) [9–11], and resistance to industrial processing [12,13]. Crucially, when postbiotics are generated from plant-derived substrates, the resulting preparations sit at the conceptual interface of postbiotic biology and phytopharmacology. Fermented herbal preparations from traditional medical systems exemplify this convergence: what is administered is no longer purely plant material but a plant–microbe co-processed system in which postbiotic metabolites mediate the therapeutic action [4,14].

A phytomedicine based framework is emerging that integrates prebiotic phytochemicals with probiotic strains to modulate gut microbiota and host health [15]. Building on this evidence, Possemiers et al. [16] have argued that the intestinal microbiome should be regarded as a separate metabolic organ with the capacity to determine the bioactivity of botanicals. Extending this position, we propose here that postbiotics generated from plant substrates can be conceptualised as *microbially matured phytomedicines*, in which the microbiome serves as an obligate biotransformation step rather than an incidental contributor. This framing has both regulatory and translational implications: it positions controlled in vitro fermentation of plant materials as a route to standardised phytomedicine preparations whose active pharmacological mediators are postbiotic in nature, and it dissolves the historical source-based distinction between phytomedicines and microbial products into a unified pharmacological category. Combinations of prebiotic plant substrates with the postbiotic preparations they generate termed here *pre-postbiotics* offer the additional advantage of synergistic bioactivity [17–19], since the residual prebiotic substrate continues to support microbial activity in vivo while the postbiotic component delivers immediate, viability-independent pharmacological effects.

Among candidate plant substrates for microbially matured phytomedicine, sesame (*Sesamum indicum* L.) hull is particularly attractive. Sesame hull is an agro-industrial byproduct that typically accounts for 15–18% of total seed mass and is currently discarded

or used as low-value animal feed [20,21], despite a complex carbohydrate composition rich in pectic and hemicellulosic polysaccharides [22]. Oligosaccharides from agro-industrial residues such as cereal (wheat, rye, barley straw) are established prebiotic substrates with known capacity to selectively stimulate beneficial gut bacteria [23–25]. Plant cell-wall polysaccharides are complex carbohydrates composed of monosaccharides linked by glycosidic bonds, principally cellulose, hemicellulose, and pectin [26]. Pectic and hemicellulosic polysaccharides and their derived oligosaccharides exhibit functional properties prebiotic, antioxidant, immunomodulatory that depend on monomer composition, degree of polymerisation (DP), and glycosidic linkage pattern [22,27]. In the context of phytomedicine, these structural features determine the substrate specificity of microbial fermentation and therefore the chemical identity of the resulting bioactive postbiotic metabolites. Differential fermentation of structurally distinct plant cell-wall oligosaccharides preparations by different *Lactobacillus* strains can therefore be expected to generate phytomedicine preparations with differential pharmacological profiles a hypothesis that is directly testable.

In this study, we test this hypothesis by fermenting three structurally distinct sesame-hull oligosaccharide extracts with three *Lactobacillus* strains (*L. acidophilus*, *L. casei*, *L. paracasei*). Under the revised taxonomy proposed by Zheng et al. [28], the latter two species are now classified within the genus *Lacticaseibacillus*; for consistency with the prior literature, the historical genus name is retained throughout. In a previous study, Miran et al. characterised the structural features of extracted sesame hull cell wall polysaccharides and demonstrated that controlled enzymatic hydrolysis can release oligosaccharides, particularly (arabino-)xylooligosaccharides ((A)XOS) [20]. These compounds were thoroughly characterised and may exhibit functional properties based on their structural features. In this study, we select the oligosaccharide obtainment procedures based on their structure, as determined by previous results, and investigate their prebiotic activity. The fermented oligosaccharides are then used to produce pre-postbiotics. To date, the prebiotic activity and postbiotic production, as well as the phytomedicinal activity of these fermented carbohydrates from sesame hulls, have not been previously explored. The resulting heat inactivated pre-postbiotic preparations are profiled for the phytomedicine-relevant pharmacological activities most consistently associated with plant-derived therapeutics: antioxidant capacity (by ABTS and FRAP), inhibition of metabolic enzymes implicated in obesity and type 2 diabetes (pancreatic lipase, α-amylase, α-glucosidase), and anti-*Helicobacter pylori* urease activity. The lead preparation is then incorporated into a barley malt (*Hordeum vulgare* L.) beverage as a translational model of a functional phytomedicine product. Together, these experiments characterise sesame hull-derived pre-postbiotics as a candidate microbially matured phytomedicine and provide a methodological template for the controlled generation of standardised phytomedicine preparations from agro-industrial plant residues.

2. Materials and Methods

2.1. Plant Substrate, Cell-Wall Polysaccharide Extraction, and Oligosaccharides Production

2.1.1. Plant Substrate

White sesame (*Sesamum indicum* L.) hull waste was obtained from Oghab Halva Company (Shahriar, Iran). At the facility, hulls were separated from seeds using a wet salt-water process, then filtered and washed until the electrical conductivity of the soaking distilled water reached 50 µS/cm. Hulls were oven-dried at 45 °C, ground, and passed through 500 µm stainless-steel sieves [20].

2.1.2. Cell-Wall Polysaccharide Extraction

Polysaccharides were extracted by alkaline (ALK) or enzymatic (Celluclast, C6h) procedures based on Miran et al. [20]. Briefly, for ALK extraction, dried hulls (50 g) were

treated with 1500 mL of 2% NaOH shaken in a water bath at 80 °C for 90 min, filtered, acidified to pH 5 with acetic acid, precipitated with 1500 mL of 96% ethanol overnight, centrifuged at 12,000× *g* for 15 min at 10 °C, washed five times with 50% ethanol, and freeze-dried (denoted ALK(P)). For enzymatic extraction, hulls (50 g) were incubated with 500 mL of sodium acetate buffer (0.05 M, pH 5) and Celluclast® 1.5 L (0.3 mL/g sesame hull) at 50 °C with constant shaking (200 rpm) for 6 h. Following this, the mixture was then heat-inactivated, filtered, precipitated with 96% ethanol overnight, centrifuged, washed three times with 70% ethanol, and freeze-dried (denoted C6h(P)). The methods were chosen from a wider range of techniques previously examined by Miran et al. [20], focusing on extraction yield, structural characteristics, and the desired oligosaccharides (see Section 2.1.3).

2.1.3. Oligosaccharides Production by Ultraflo Hydrolysis

Each polysaccharide preparation (3 g) was hydrolysed with Ultraflo® L (1.4 mL/g of extracted polysaccharides) in 150 mL of sodium acetate buffer (0.05 M, pH 5) at 40 °C, with constant shaking (200 rpm) for either 1 h or 6 h. The reaction was then heat-inactivated and freeze-dried [20]. The resulting oligosaccharides preparations are denoted *a* + 6 (ALK(P) hydrolysed for 6 h), *c* + 1 (C6h(P) hydrolysed for 1 h), and *c* + 6 (C6h(P) hydrolysed for 6 h). In this study, we selected the oligosaccharides producing methods (including enzyme type, dosage, incubation time and temperature) based on previous findings. The selection process was conducted from a broader array of techniques examined by Miran et al. [20] and was based on the structural features of these oligosaccharides, in conjunction with a preliminary screening of their potential to promote probiotic growth.

2.1.4. Structural Characterisation of Oligosaccharides

Structural characterisation of the polysaccharide and oligosaccharides preparations was previously reported by Miran et al. [20]. In summary, ALK(P) was composed of 34.6% galacturonic acid, 28.5% xylose, 25.0% arabinose, 5.1% galactose, 4.2% rhamnose, and minor amounts of glucose, glucuronic acid, ferulic acid, fucose, and mannose; C6h(P) was rich in pectin (54.9% galacturonic acid) with substantial arabinoxylan (13.6% xylose, 9.1% arabinose). For the AXOS preparations, molecular weight by HPSEC-ELSD ranged 29.7–0.2 kDa for *a* + 6, 38.0–0.2 kDa for *c* + 1, and 36.3–0.2 kDa for *c* + 6. MALDI-TOF-MS analysis confirmed AXOS as the predominant oligosaccharide species in *a* + 6 (77%, DP 2–13), *c* + 1 (~50%, DP 3–22, with one ferulic-acid-substituted AXOS), and *c* + 6 (similar series to *c* + 1 with lower total intensity).

2.2. Lactobacillus Fermentation and Postbiotic Preparation

Three *Lactobacillus* strains were used as probiotics: *L. acidophilus*, *L. casei*, and *L. paracasei* (under the Zheng et al. [28] taxonomy, the latter two are *Lacticaseibacillus casei* and *Lacticaseibacillus paracasei*). These *Lactobacillus* strains were obtained from the culture collection of the Bioprocess Engineering Laboratory at the Department of Food Science and Engineering, University of Tehran, Iran. They were activated through two successive passages in sterilised De Man, Rogosa and Sharpe (MRS) broth from Merck, incubated at 37 °C for 12 h. Following this, 40% (*v*/*v*) sterilised glycerol was added to the culture, which was then stored at −20 °C in sterile screw-cap tubes.

One mL of activated culture in liquid MRS medium (10^7–10^8 CFU) was inoculated into 8 mL of sterile MRS containing 1 mL of pasteurised (80 °C, 5 min) oligosaccharides solution (1 mg/mL in distilled water). Controls received 1 mL of distilled water in place of oligosaccharides. Cultures were incubated at 37 °C for 24 h [29–31]. After incubation, samples were centrifuged at 13,570× *g* for 5 min at 4 °C, and the supernatant filtered through a 0.45 µm membrane. To inactivate the cells, the cell pellet (cells) were heated at 95 °C for 15 min, then recombined with the supernatant, and freeze-dried to yield the

final pre-postbiotic powder. Bacterial cell counts were determined by serial dilution plate counting [32] and expressed as log CFU/mL.

2.3. Pharmacological Profiling of Pre-Postbiotic Preparations

2.3.1. ABTS Radical Scavenging Assay

ABTS•$^+$ (2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid)) radical scavenging activity was determined by a modified Malunga & Beta method [33]. Pre-postbiotic samples (1 and 3 mg/mL in pH 8 phosphate buffer) were mixed with 0.9 mL of ABTS•$^+$ reagent in 2 mL tubes, vortexed, and incubated in the dark for 30 min. Absorbance was read at 734 nm using a UV–Vis spectrophotometer (Cecil Instruments Ltd., Cambridge, UK). Ascorbic acid and gallic acid (1 and 3 mg/mL) served as positive controls [29].

$$\text{ABTS scavenging (\%)} = [(\text{A_blank} - \text{A_sample})/\text{A_blank}] \times 100 \quad (1)$$

where A_blank is the absorbance of the ABTS•$^+$ reagent mixed with buffer (no sample), and A_sample is the absorbance of the ABTS•$^+$ reagent mixed with the test sample.

2.3.2. Ferric Reducing Antioxidant Power (FRAP) Assay

FRAP was determined by the method of Gullón et al. [34]. The FRAP reagent was 45 mL of 300 mM acetate buffer (pH 3.6), 4.5 mL of 10 mM 2,4,6-tripyridyl-s-triazine (TPTZ) in 40 mM HCl, and 4.5 mL of 20 mM $FeCl_3 \cdot 6H_2O$ in distilled water. Samples (0.1 mL of 3 and 5 mg/mL in pH 8 phosphate buffer) were combined with 3 mL of FRAP reagent, vortexed, and incubated in the dark for 4 min. Absorbance was read at 593 nm using a UV–Vis spectrophotometer (Cecil Instruments Ltd., Cambridge, UK). $FeSO_4 \cdot 7H_2O$ (0.02 mg/mL) served as the positive control.

$$\text{Ferric reducing capacity (\%)} = (\text{A_sample} - \text{A_blank})/(\text{A_positive} - \text{A_blank}) \times 100 \quad (2)$$

where A_blank is FRAP reagent plus buffer, A_positive is FRAP reagent plus $FeSO_4 \cdot 7H_2O$ (positive control), and A_sample is FRAP reagent plus test sample.

2.3.3. Pancreatic Lipase Inhibition

Samples (3 mg/mL in pH 8 phosphate buffer; 40 µL) were mixed with 20 µL of substrate (10 mM *p*-nitrophenylbutyrate in DMSO) and 40 µL of porcine pancreatic lipase (2.5 mg/mL in 0.1 M phosphate buffer, pH 8). After 20 min at 37 °C, absorbance was measured at 405 nm using an ELISA reader (Expert 96, Biotech, Miami, FL, USA). Orlistat (50 µM) served as the positive reference [35].

$$\text{Lipase inhibition (\%)} = [(\text{A_control} - \text{A_sample})/\text{A_control}] \times 100 \quad (3)$$

where A_control is the absorbance of the reaction mixture without the sample, and A_sample is the absorbance of the reaction mixture containing the pre-postbiotic sample.

2.3.4. α-Amylase Inhibition

Samples (80 µL of 3 mg/mL in pH 8 phosphate buffer) were combined with 80 µL of starch substrate (2 mg/mL in 0.1 M phosphate buffer, pH 6.5) and 80 µL of α-amylase (0.6 mg/mL in 0.1 M phosphate buffer, pH 6.5). After 20 min at 37 °C, the reaction was stopped with 400 µL of 3,5-dinitrosalicylic acid (DNS) reagent containing 1% DNS in alkaline sodium potassium tartrate solution and heated at 100 °C for 5 min. Absorbance was read at 540 nm using an ELISA reader (Expert 96, Biotech, Miami, FL, USA). Acarbose (1 mg/mL) served as the positive control [27].

$$\alpha\text{-Amylase inhibition (\%)} = [(\text{A_control} - \text{A_sample})/\text{A_control}] \times 100 \quad (4)$$

where A_control is the absorbance of the enzyme reaction without inhibitor and A_sample is the absorbance with the test sample.

2.3.5. α-Glucosidase Inhibition

Samples (100 µL of 3 mg/mL in pH 8 phosphate buffer) were preincubated for 10 min at 37 °C with 100 µL of α-glucosidase (0.5 U/mL in 100 mM phosphate buffer, pH 6.9). 100 µL of 5 mM *p*-nitrophenyl-α-D-glucopyranoside (pNPG) was added, and absorbance was read after 20 min at 37 °C using an ELISA reader (Expert 96, Biotech, Miami, FL, USA). Acarbose (1 mg/mL) served as the positive control [27].

$$\alpha\text{-Glucosidase inhibition (\%)} = [(\text{A_control} - \text{A_sample})/\text{A_control}] \times 100 \quad (5)$$

where A_control is the absorbance of the reaction without inhibitor and A_sample is the absorbance with the test sample.

2.3.6. Anti-*Helicobacter pylori* Activity (Urease Inhibition)

Anti-*H. pylori* activity was assessed by urease inhibition via Berthelot ammonium release [36]. Samples (100 µL of 3 mg/mL in pH 8 phosphate buffer) were combined with 850 µL of 30 mM urea and 50 µL of urease (1 mg/mL in 100 mM phosphate buffer, pH 7.6) and incubated at 37 °C for 30 min. A 100 µL aliquot was added sequentially to 500 µL of solution A (4.47 g salicylic acid, 2.5 g NaOH, 20 mg sodium nitroprusside in 50 mL water) and solution B (1.5 mL of 5% sodium hypochlorite solution and 0.5 g NaOH in 70 mL water), incubated at 37 °C for 30 min, and the absorbances were measured at 625 nm using an ELISA reader (Expert 96, Biotech, Miami, FL, USA).

$$\text{Urease inhibition (\%)} = [(\text{A_control} - \text{A_sample})/\text{A_control}] \times 100 \quad (6)$$

where A_control is the absorbance of the urease reaction without the test sample (buffer instead of sample) and A_sample is the absorbance in the presence of the test sample.

2.4. Translational Application: Pre-Postbiotic Barley Malt Beverage

A commercial ready-to-drink barley malt (*Hordeum vulgare* L.) beverage was used as a translational carrier. Pre-postbiotic powder derived from *c* + 6 oligosaccharide fermentation (selected on the basis of superior in vitro pharmacological profile) was added aseptically at 1, 2, and 3% *w*/*v* in the commercial ready-to-drink barley malt beverage during the final production stage, and 1% *w*/*v* was selected after preliminary screening. Final formulations were assessed for antioxidant activity (ABTS, FRAP; Sections 2.3.1 and 2.3.2), pH (calibrated meter, dried electrode in 10 mL aliquots after particles had settled), and apparent optical density at 600 nm (OD600). OD600 was measured spectrophotometrically at 600 nm against a distilled-water blank and was used as an indirect comparative indicator of beverage cloudiness and suspended particulates under standardised measurement conditions.

2.5. Sensory Evaluation

Sensory evaluation of the malt beverages was conducted with 30 panellists (*n* = 15 female, *n* = 15 male; age range 22–58 years) drawn from the Department of Food Science at the University of Tehran. Panellists rated samples on a five-point quality scale (1 = extremely poor; 5 = excellent) for taste, aroma, colour, aftertaste, mouthfeel, and overall acceptability. Four samples (one blank and three pre-postbiotic-supplemented) were presented in randomised order under a three-digit code, with palate-cleansing water provided between samples.

2.6. Statistical Analysis

Experiments were conducted in triplicate ($n = 3$) and results expressed as mean ± standard deviation. Data were analysed by one-way ANOVA with Duncan's multiple range test at $p < 0.05$ using Minitab 20 (Minitab LLC, State College, PA, USA). Equivalent results were obtained with Tukey's HSD.

2.7. Ethics Statement

All sensory evaluation procedures were approved by the Research Ethics Committee of the University of Tehran (Approval No. ETHIC-202601-1665) and conducted in accordance with the 1964 Declaration of Helsinki and its later amendments. Informed consent was obtained verbally from all participants prior to evaluation.

3. Results and Discussion

3.1. Sesame-Hull Oligosaccharides as Plant-Derived Prebiotic Substrate for Lactobacilli

Phytomedicine generated by microbial biotransformation of plant material requires that the plant substrate first sustain microbial growth. We therefore confirmed that all three *Lactobacillus* species could grow on sesame-hull-derived AXOS as carbon source. After 24 h fermentation in MRS supplemented with 1 mg/mL AXOS, all strains exhibited significantly higher growth than oligosaccharide-free controls (Table 1; $p < 0.05$), demonstrating that the cell-wall oligosaccharides released from sesame hull are bioavailable to lactic-acid-fermenting microbiota and meet the operational definition of a prebiotic substrate [37]. *L. casei* supported the highest growth (Δlog CFU/mL up to 2.43), *L. paracasei* the second-highest, and *L. acidophilus* the lowest. The relative inability of *L. acidophilus* to metabolise complex AXOS structures has been previously reported [38] and likely reflects strain-specific glycoside hydrolase repertoires; the superior growth of *L. casei* may additionally relate to its higher exopolysaccharide-production capacity, which improves environmental stress tolerance [39].

Table 1. Effect of sesame-hull-derived cell-wall AXOS on growth of three *Lactobacillus* species after 24 h fermentation.

Probiotic Strain	Sample	Log CFU/mL (t = 0)	Log CFU/mL (t = 24)	Δlog CFU/mL
L. acidophilus (LA)	Control	7.970 ± 0.020	8.940 ± 0.030	0.970 ^{c}
	a + 6	8.120 ± 0.003	9.390 ± 0.004	1.270 ^{b}
	c + 1	8.120 ± 0.008	9.460 ± 0.002	1.340 ^{ab}
	c + 6	8.110 ± 0.003	9.490 ± 0.016	1.380 ^{a}
L. casei (LCa)	Control	7.100 ± 0.001	8.840 ± 0.061	1.740 ^{b}
	a + 6	7.070 ± 0.009	9.370 ± 0.010	2.300 ^{a}
	c + 1	7.170 ± 0.009	9.480 ± 0.009	2.310 ^{a}
	c + 6	7.180 ± 0.003	9.610 ± 0.020	2.430 ^{a}
L. paracasei (LPC)	Control	7.650 ± 0.004	8.840 ± 0.007	1.180 ^{c}
	a + 6	7.640 ± 0.004	9.310 ± 0.009	1.670 ^{b}
	c + 1	7.650 ± 0.016	9.540 ± 0.002	1.900 ^{a}
	c + 6	7.650 ± 0.014	9.580 ± 0.030	1.980 ^{a}

Values are mean ± SD ($n = 3$). Different superscripts (a, b, c) indicate significant differences ($p < 0.05$) within strain; comparisons were not made across strains. Sample codes: *a* + 6 = AXOS from alkaline-extracted polysaccharide hydrolysed 6 h with Ultraflo; *c* + 1/*c* + 6 = AXOS from Celluclast-extracted polysaccharide hydrolysed 1 or 6 h with Ultraflo; Control = MRS without AXOS.

Distinctions in monomeric composition and chemical structure of oligosaccharides significantly influence their prebiotic activity. Among the oligosaccharides preparations, *c* + 1 and *c* + 6 from extended Celluclast–Ultraflo hydrolysis supported the highest growth across all strains; the higher activity observed may be attributed to variations in their monomeric composition; as *c* + 1 and *c* + 6 have different compositions compared to *a* + 6, consistent with the well-established principle that distinctions in monomeric composition modulate oligosaccharides fermentability and the consequent yield of bioactive postbiotic metabolites including short-chain fatty acids (SCFAs) [23,40]. The differential growth observed across structurally distinct AXOS preparations therefore predicts differential pharmacological profiles for the resulting postbiotics—the central testable hypothesis of the present phytomedicine framework. Comparable strain × substrate interactions have been documented for xylo-oligosaccharides from banana pseudostem [41] and for arabinoxylan hydrolysates from wheat and rye [25], confirming that AXOS-driven prebiotic activity is a general property of plant-derived cell-wall oligosaccharides.

3.2. Antioxidant Bioactivity: ABTS and FRAP

Antioxidant activity is one of the most extensively documented pharmacological effects of plant-derived therapeutics, attributable largely to polyphenols, phenolic acids, and structurally related microbial metabolites [2,14,42]. In the present plant–microbe system, fermentation of AXOS substrates by gut-relevant *Lactobacillus* species generates SCFAs (acetate, propionate, butyrate), bioactive peptides, and organic acids that contribute to antioxidant capacity [43–45]. We assessed antioxidant activity by two complementary assays.

In the ABTS assay (Table 2), antioxidant activity increased dose-dependently across all preparations (1 → 3 mg/mL; $p < 0.05$), and the AXOS-supplemented postbiotics consistently outperformed the AXOS-free controls. *L. paracasei* generated the most potent antioxidant postbiotics: at 3 mg/mL, LPC(*c* + 6) exhibited 87.02% radical scavenging—within the range typical of high-potency plant antioxidant extracts. The corresponding FRAP data (Table 3) confirmed concentration-dependent ferric-reducing capacity, with LPC(*c* + 1) the most active at 5 mg/mL (65.85%). Differences between ABTS and FRAP results across strain × substrate combinations reflect mechanistic differences between the assays (radical scavenging vs. single-electron reduction) and underscore the value of orthogonal antioxidant measurements when characterising candidate phytomedicine preparations [44].

Table 2. ABTS radical-scavenging activity of pre-postbiotic preparations from sesame-hull AXOS fermentation.

Sample	Antioxidant Activity (%) at 1 mg/mL	Antioxidant Activity (%) at 3 mg/mL
LA (control)	8.82 ± 1.31 ^{c}	63.70 ± 0.24 ^{c}
LA(*a* + 6)	17.16 ± 0.50 ^{b}	70.91 ± 0.56 ^{b}
LA(*c* + 1)	26.98 ± 2.23 ^{a}	80.21 ± 1.52 ^{a}
LA(*c* + 6)	29.79 ± 0.55 ^{a}	81.65 ± 2.16 ^{a}
LPC (control)	32.90 ± 0.83 ^{c}	72.43 ± 0.96 ^{c}
LPC(*a* + 6)	41.60 ± 0.95 ^{b}	82.61 ± 2.64 ^{ab}
LPC(*c* + 1)	41.36 ± 0.95 ^{b}	80.29 ± 0.16 ^{b}
LPC(*c* + 6)	60.19 ± 0.72 ^{a}	87.02 ± 0.16 ^{a}
LCa (control)	30.39 ± 1.11 ^{b}	69.31 ± 0.24 ^{c}
LCa(*a* + 6)	37.54 ± 1.91 ^{a}	73.08 ± 0.32 ^{b}

Table 2. *Cont.*

Sample	Antioxidant Activity (%) at 1 mg/mL	Antioxidant Activity (%) at 3 mg/mL
LCa(*c* + 1)	40.64 ± 2.31 [a]	72.92 ± 1.60 [b]
LCa(*c* + 6)	41.00 ± 0.60 [a]	81.25 ± 0.51 [a]

Values are mean ± SD (n = 3). Different superscripts indicate significant differences ($p < 0.05$) within column and the strain. LA, *L. acidophilus*; LCa, *L. casei*; LPC, *L. paracasei*. Suffixes (*a* + 6), (*c* + 1), (*c* + 6) denote the AXOS substrate; "control" denotes postbiotic produced without AXOS.

Mechanistically, our findings are consistent with prior reports that synbiotic combinations of *Lactobacillus* species with structurally diverse oligosaccharides synergistically enhance antioxidant activity beyond that of either component alone [45], and that polysaccharides released from heat-inactivated *L. acidophilus* cell walls themselves possess antioxidant and anticancer activity [46]. From the phytomedicine perspective, these data demonstrate that a plant-derived substrate (sesame-hull AXOS) is converted by *Lactobacillus* fermentation into a postbiotic preparation whose antioxidant activity reflects both the phenolic content of the original plant material (residual ferulic acid and related substituents on AXOS [20]) and the microbial metabolites generated during fermentation. The microbially matured product is, by both compositional and functional criteria, more pharmacologically active than the parent plant fraction.

Table 3. Ferric-reducing antioxidant power (FRAP) of pre-postbiotic preparations from sesame-hull AXOS fermentation.

Sample	Reducing Activity (%) at 3 mg/mL	Reducing Activity (%) at 5 mg/mL
LA (control)	5.31 ± 0.94 [b]	18.62 ± 2.32 [b]
LA(*a* + 6)	27.51 ± 2.27 [a]	51.40 ± 1.51 [a]
LA(*c* + 1)	28.65 ± 3.41 [a]	51.28 ± 2.70 [a]
LA(*c* + 6)	39.15 ± 1.06 [a]	56.90 ± 1.97 [a]
LPC (control)	12.90 ± 0.18 [c]	19.58 ± 1.27 [c]
LPC(*a* + 6)	25.81 ± 2.08 [b]	49.77 ± 2.55 [b]
LPC(*c* + 1)	40.23 ± 2.46 [a]	65.85 ± 2.32 [a]
LPC(*c* + 6)	44.78 ± 2.08 [a]	56.76 ± 0.58 [ab]
LCa (control)	9.11 ± 2.46 [b]	17.83 ± 1.16 [b]
LCa(*a* + 6)	28.27 ± 0.75 [a]	55.24 ± 1.33 [a]
LCa(*c* + 1)	32.26 ± 0.18 [a]	47.55 ± 0.11 [a]
LCa(*c* + 6)	30.93 ± 1.51 [a]	46.97 ± 0.81 [a]

Values are mean ± SD (n = 3); see Table 2 for sample-code definitions. Different superscripts indicate significant differences ($p < 0.05$) within column and the strain.

3.3. Anti-Obesity Pharmacology: Pancreatic Lipase Inhibition

Pancreatic lipase hydrolyses 50–70% of dietary triglycerides and is a clinically validated target for anti-obesity phytomedicines [47]—orlistat, the only FDA-approved long-term anti-obesity lipase inhibitor, is itself a microbial product (a hydrogenated derivative of lipstatin from *Streptomyces toxytricini*). All three *Lactobacillus*-derived pre-postbiotics significantly inhibited pancreatic lipase relative to AXOS-free controls (Figure 1; $p < 0.05$). LCa(*a* + 6) was the most potent inhibitor at 47.82%, an activity comparable to several plant phenolic phytomedicine preparations [48]. The strain-specificity of the response—*L. casei*-derived postbiotics outperformed both *L. acidophilus*- and *L. paracasei*-derived preparations—

supports the hypothesis that the microbial enzymatic complement that processes a plant substrate determines the pharmacological identity of the resulting phytomedicine.

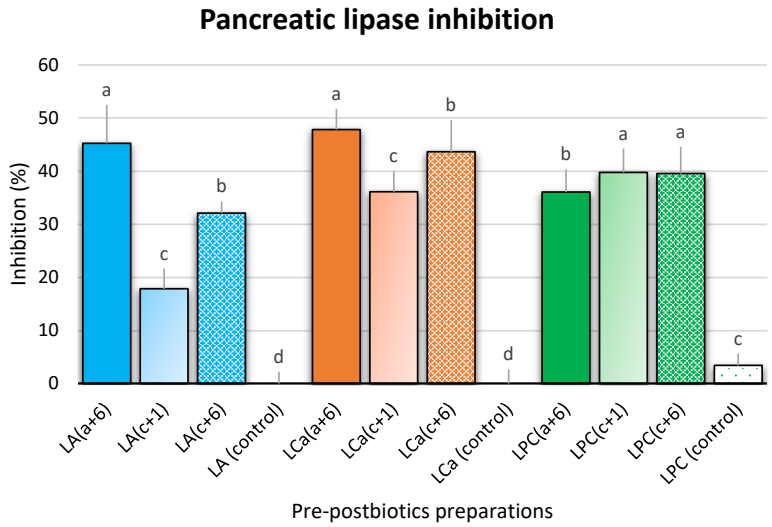


Figure 1. Pancreatic lipase inhibition (%) of pre-postbiotic preparations (3 mg/mL in pH 8 phosphate buffer) from sesame-hull AXOS fermentation. Values are mean ± SD (n = 3). Different lowercase letters denote $p < 0.05$ within strain. LA, *L. acidophilus*; LCa, *L. casei*; LPC, *L. paracasei*. Suffixes (a + 6), (c + 1), (c + 6) denote the oligosaccharides substrate; "control" denotes postbiotic produced without AXOS.

Mechanistically, *L. casei*-derived postbiotics have been reported to allosterically inhibit pancreatic lipase, bile-salt-stimulated lipase, and phospholipase A_2 via specific bioactive metabolites including 8-C-glucosylorobol [48], although whether such metabolites are present in our AXOS-fermented preparations remains to be confirmed by targeted metabolomics. More broadly, the physicochemical properties of the bacterial cell-wall surface (notably hydrophobicity) enhance adhesion to lipid substrates and may sterically interfere with lipase access [49], while fermentation-derived peptides and polysaccharides can compete with or inhibit the catalytic site [50]. From the phytomedicine perspective, these multiple convergent mechanisms reinforce the value of microbially matured postbiotic preparations as multi-target anti-obesity therapeutics, complementary to single-component plant phenolics.

3.4. Anti-Diabetic Pharmacology: α-Amylase Inhibition

α-Amylase and α-glucosidase are key targets for the anti-hyperglycaemic action of many plant-derived phytomedicines—acarbose, miglitol, and voglibose, the principal pharmaceutical α-glucosidase inhibitors, derived from microbial natural products and related genera, illustrating that microbially derived therapeutics have long served as anti-diabetic agents [51]. All three *Lactobacillus*-derived pre-postbiotics inhibited α-amylase activity at 3 mg/mL (Figure 2). LA(a + 6) was the most potent inhibitor at 43.67%, with LPC(c + 1) the least at 15.32%; AXOS-free controls showed no detectable inhibition. The pattern—maximal inhibition for the alkaline-extracted AXOS substrate (a + 6) in *L. acidophilus* and *L. casei*, and for c + 6 in *L. paracasei*—again reflects the strain × substrate dependence of the resulting phytomedicine pharmacology.

Suwanangul et al. [52] reported that probiotic metabolites including organic acids, SCFAs, peptides, and exopolysaccharides interfere with α-amylase and α-glucosidase active sites, potentially via substrate mimicry or catalytic-residue binding; *Bacillus coagulans* metabolites display similarly potent inhibitory activity. From the phytomedicine perspective, the implication is significant: a plant-derived substrate (sesame-hull AXOS) that

has no inherent α-amylase inhibitory activity in its native form acquires substantial pharmacological activity—comparable to many plant phenolic extracts—following microbial biotransformation. This is the canonical signature of a microbially matured phytomedicine.

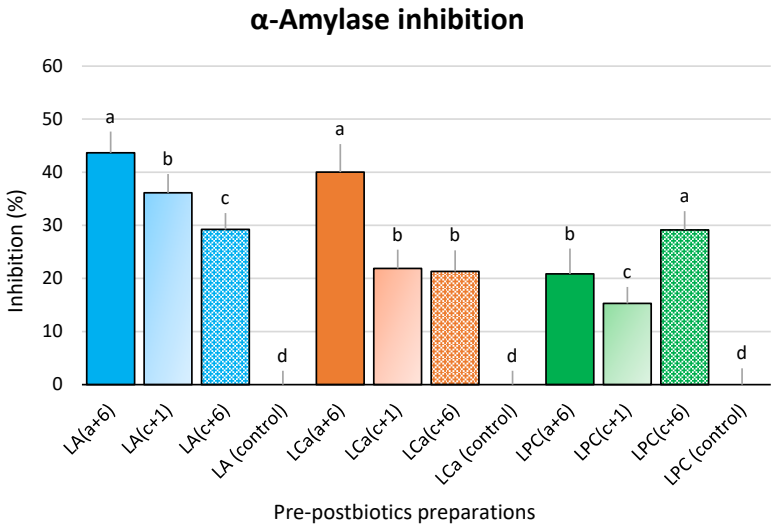


Figure 2. α-Amylase inhibition (%) of pre-postbiotic preparations (3 mg/mL in pH 8 phosphate buffer). Values are mean ± SD (n = 3). Different lowercase letters denote $p < 0.05$ within strain.

3.5. Anti-Diabetic Pharmacology: α-Glucosidase Inhibition

All pre-postbiotic preparations inhibited α-glucosidase, with LCa(*a* + 6) the most potent at 52.14% and LCa(*c* + 6) the weakest at 29.60% (Figure 3). AXOS-free controls were inactive. As with pancreatic lipase, *L. casei*-derived postbiotics from the alkaline-extracted AXOS substrate were the lead inhibitors. *L. casei* produces higher levels of exopolysaccharides than *L. acidophilus* [53,54]; given that exopolysaccharides have been linked to α-glucosidase inhibition, the increased inhibitory activity of *L. casei*-derived preparations is mechanistically plausible. The simultaneous lipase- and glucosidase-inhibitory activity of LCa(*a* + 6) is particularly relevant from the phytomedicine perspective: dual-target pharmacology against complementary metabolic enzymes is a hallmark of clinically valuable plant-derived therapeutics for metabolic syndrome.

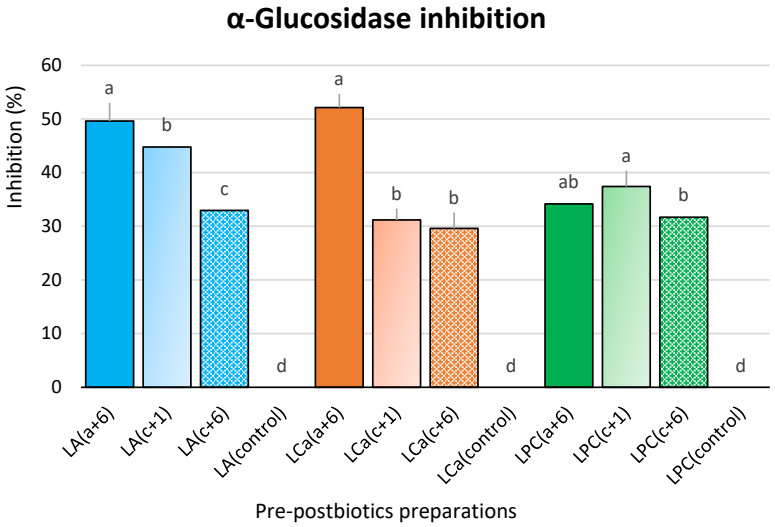


Figure 3. α-Glucosidase inhibition (%) of pre-postbiotic preparations (3 mg/mL in pH 8 phosphate buffer). Values are mean ± SD (n = 3). Different lowercase letters denote $p < 0.05$ within strain.

3.6. Anti-Helicobacter pylori Urease Activity

Plant-derived antibacterial phytomedicines have a long history of use against *H. pylori*, with the urease enzyme—essential for *H. pylori* colonisation of the gastric mucosa via local pH neutralisation—a primary pharmacological target [55]. We assessed urease inhibition at 3 mg/mL (Figure 4). Only preparations with detectable urease inhibition are shown. LPC(*c* + 6) exhibited the highest activity at 20.66%, significantly above all other preparations; AXOS-free controls were inactive. *L. paracasei* has previously been reported to display strong anti-*H. pylori* activity attributed to bacteriocin and organic-acid production [56,57], and strains with higher antioxidant activity tend to display stronger anti-*H. pylori* properties [58]. The dual antioxidant and anti-*H. pylori* activity of LPC(*c* + 6) is therefore mechanistically coherent and therapeutically attractive: the same preparation simultaneously addresses gastric oxidative stress and the principal pathogen of chronic gastritis and gastric ulcer disease, in line with the multi-target therapeutic profile expected of a microbially matured phytomedicine.

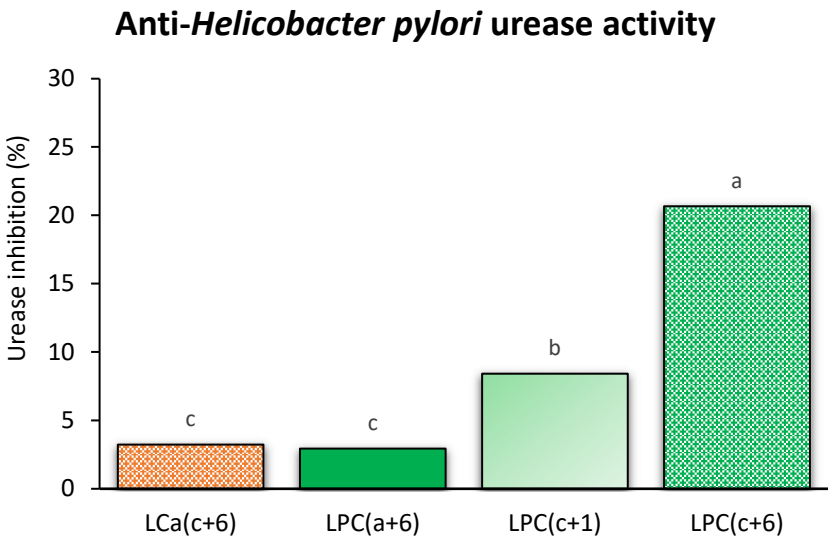


Figure 4. Anti-*Helicobacter pylori* urease inhibition (%) (3 mg/mL in pH 8 phosphate buffer). Values are mean ± SD (*n* = 3). Different lowercase letters denote $p < 0.05$.

3.7. Translational Phytomedicine: Antioxidant Activity in Pre-Postbiotic Barley Malt Beverage

A central challenge in phytomedicine development is translating in vitro pharmacological activity into a stable, palatable delivery vehicle. We selected barley malt (*Hordeum vulgare* L.) as a translational carrier: barley is itself a plant material with intrinsic phytomedicinal heritage (recognised antioxidant content enhanced through malting [59]), and barley malt beverages are widely consumer-accepted. Pre-postbiotic powder from *c* + 6 AXOS fermentation—selected on the basis of its in vitro pharmacological profile—was incorporated at 1% *w*/*v* (higher concentrations produced unacceptable excessive cloudiness and discolouration).

ABTS-measured antioxidant activity of the supplemented beverages increased significantly ($p < 0.05$) by approximately 20% relative to the unsupplemented blank, with LCa(*c* + 6) showing the strongest enhancement (67.75%; Figure 5). This represents a clinically relevant improvement in radical-scavenging capacity in a real food matrix, achieved using a 1% *w*/*v* inclusion of a microbially matured phytomedicine derived entirely from agricultural plant byproducts. The translational case for sesame-hull-derived pre-postbiotic phytomedicines is therefore supported by both component-level pharmacology (Sections 3.2–3.6) and product-level functional behaviour.

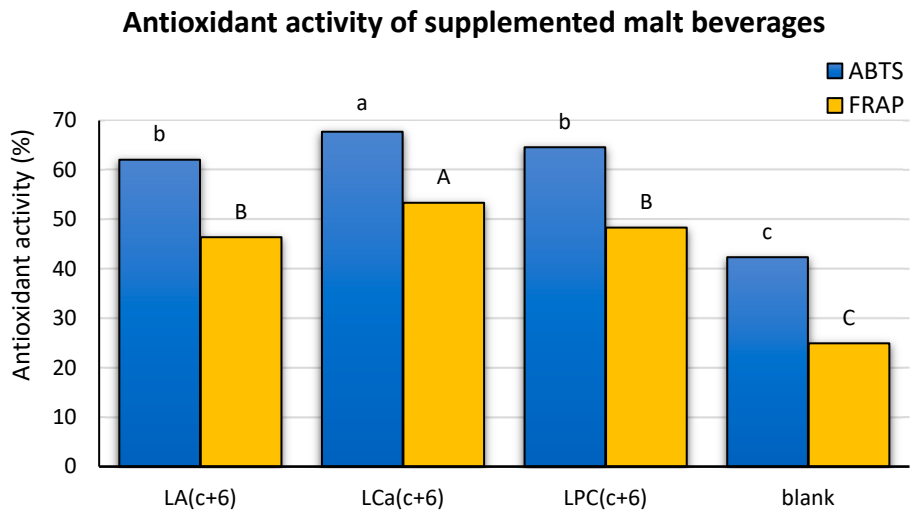


Figure 5. ABTS and FRAP antioxidant activity of barley malt beverages supplemented with 1% *w*/*v* pre-postbiotic powder. "Blank" denotes unsupplemented beverage. Values are mean ± SD (n = 3); different letters denote $p < 0.05$ within assay.

3.8. Physicochemical Characteristics of the Pre-Postbiotic Beverage

Beverage pH and optical density at 600 nm (OD600) were evaluated as physicochemical characteristics relevant to beverage quality and visual appearance (Table 4) [60]. All formulations exhibited an acidic pH (4.57–4.62), characteristic of fermented and fermentation-derived beverages and consistent with reduced spoilage risk. Slight pH differences between strains likely reflect residual organic acids carried over from the fermentation process rather than de novo acid production by the inactivated cells. OD600 values differed significantly among formulations, with LCa(*c* + 6) showing the highest value (2.760) and LPC(*c* + 6) the lowest (2.260). OD600 was used here as a comparative optical indicator of beverage cloudiness and suspended particulates under standardised measurement conditions [60]. The observed differences may reflect variation in suspended biomass and strain-dependent exopolysaccharide production.

Table 4. pH and apparent optical density (OD600) of barley malt beverages supplemented with pre-postbiotic preparations from *c* + 6 oligosaccharides fermentation.

Sample	pH	OD600
LA(*c* + 6)	4.570 ± 0.005 ^{c}	2.370 ± 0.002 ^{b}
LCa(*c* + 6)	4.580 ± 0.006 ^{bc}	2.760 ± 0.020 ^{a}
LPC(*c* + 6)	4.620 ± 0.001 ^{a}	2.260 ± 0.005 ^{c}
Blank	4.600 ± 0.005 ^{b}	1.230 ± 0.001 ^{d}

Values are mean ± SD (n = 3). Different superscripts denote $p < 0.05$ within column. "Blank" denotes unsupplemented beverage.

3.9. Sensory Acceptability

Consumer acceptance is essential for any plant-derived therapeutic delivered as a beverage. Sensory evaluation by an untrained panel of thirty adults from the Department of Food Science at the University of Tehran revealed no significant differences between formulations in flavour, odour, or aftertaste (Table 5). The unsupplemented blank achieved the highest scores for colour, mouthfeel, and overall acceptance, while among the supplemented formulations LA(*c* + 6) and LCa(*c* + 6) performed best. These results indicate that pre-postbiotic supplementation can be perceived organoleptically (most clearly in colour and mouthfeel), and that further formulation work—lower inclusion rates, encapsulation, masking agents—would likely be required to optimise consumer acceptance for commercial

deployment. Importantly, no formulation generated unacceptable sensory characteristics; the lead phytomedicine preparations remained within the acceptable consumer envelope.

Table 5. Sensory evaluation of barley malt beverages supplemented with pre-postbiotic preparations.

Sensory Attribute	LA(*c* + 6)	LCa(*c* + 6)	LPC(*c* + 6)	Blank
Flavour	4.50 ± 0.42 ^{a}	4.35 ± 0.39 ^{a}	4.00 ± 0.50 ^{a}	4.48 ± 0.46 ^{a}
Odour	5.00 ± 0.00 ^{a}	5.00 ± 0.00 ^{a}	5.00 ± 0.00 ^{a}	5.00 ± 0.00 ^{a}
Colour	3.80 ± 0.50 ^{b}	3.48 ± 0.53 ^{bc}	3.18 ± 0.44 ^{c}	4.68 ± 0.39 ^{a}
Mouthfeel	4.03 ± 0.40 ^{b}	3.88 ± 0.41 ^{b}	3.40 ± 0.52 ^{c}	5.00 ± 0.00 ^{a}
Aftertaste	4.58 ± 0.41 ^{a}	4.45 ± 0.35 ^{a}	4.20 ± 0.31 ^{a}	4.93 ± 0.17 ^{a}
Overall acceptance	4.30 ± 0.23 ^{b}	4.08 ± 0.33 ^{b}	3.68 ± 0.43 ^{c}	4.78 ± 0.38 ^{a}

Values are mean ± SD (n = 30). Different superscripts denote $p < 0.05$ within row. Five-point quality scale (1 = extremely poor; 5 = excellent).

3.10. Implications for Microbially Matured Phytomedicine and Functional-Beverage Development

Taken together, these data establish sesame-hull-derived pre-postbiotics as a candidate microbially matured phytomedicine with multi-target pharmacology spanning oxidative stress, dietary lipid digestion, carbohydrate digestion, and antibacterial activity against the principal pathogen of chronic gastritis. Three implications are worth emphasising. First, the pharmacological identity of the preparation is determined jointly by the plant substrate (here, sesame-hull AXOS) and the fermenting microorganism (here, *Lactobacillus* species), and is therefore tunable by selection of either component. This is precisely the design space available for deliberate phytomedicine engineering. Second, the lead preparations approach the pharmacological potency of single-component plant phytomedicines, despite being generated from material that is currently discarded as agro-industrial waste—offering a sustainable route to standardised phytomedicine ingredients. Third, the translational pathway demonstrated here (in vitro fermentation → freeze-dried postbiotic → incorporation into a plant-based beverage carrier) provides a manufacturing template that is compatible with existing food-industry infrastructure.

The global functional-beverage market was valued at over USD 208 billion in 2024 and continues to grow at a compound annual rate of approximately 7.5% [61]. Phytomedicine-positioned products within this market—those whose health claims rest on standardised, mechanistically characterised plant-derived bioactives—face both the greatest commercial opportunity and the most demanding regulatory scrutiny. The framework demonstrated here, in which a defined plant substrate is converted by defined microorganisms into a chemically and biologically characterised postbiotic preparation, is well aligned with the type of standardisation that regulators increasingly require for plant-based therapeutics [61,62]. Compliance with labelling guidelines and health-claim regulations, including ISAPP-aligned definitions of postbiotic preparations, will be essential for translation to clinical use.

3.11. Limitations

Several limitations should be considered. First, the present study did not include direct metabolomic profiling (LC-MS/MS, NMR) of the postbiotic preparations; the specific bioactive metabolites responsible for the antioxidant and enzyme-inhibitory activities therefore remain to be definitively identified. Second, the heat-killed cell biomass and the cell-free supernatant fractions were not characterised separately; the relative pharmacological contributions of cell-wall-associated and metabolite-associated activity cannot be partitioned from these data. Third, in vitro and in vivo digestion models were not employed; whether the inhibitory activities observed in buffer translate to post-prandial efficacy requires further investigation. Fourth, the sensory panel was small and not naïve,

and beverage shelf-life stability was not assessed across storage conditions both critical for commercial development. Finally, although in vitro pharmacology supports a phytomedicine framing, in vivo pharmacokinetic, toxicological, and clinical studies will be required before any therapeutic claim can be established.

4. Conclusions

Controlled fermentation of plant-derived oligosaccharides from sesame hulls using *Lactobacillus* strains produces heat-stable, freeze-dried pre-postbiotic preparations with multiple pharmacological targets relevant to phytomedicine. These preparations demonstrated enhanced antioxidant activity, achieving up to 87.02% ABTS scavenging. Furthermore, the preparations exhibited inhibitory effects against key enzymes linked to obesity and type 2 diabetes, including pancreatic lipase (47.82% inhibition), α-glucosidase (52.14% inhibition), and α-amylase (43.67% inhibition). Additionally, the preparations inhibited *Helicobacter pylori* urease by 20.66%. Consequently, the fermentation of sesame hull-derived oligosaccharides in this framework resulted in a microbially matured phytomedicine. The pharmacological activity was found to be strain- and substrate-dependent: preparations obtained by *L. casei* and oligosaccharides from alkaline-extracted extended-hydrolysis were identified as the leading anti-metabolic syndrome phytomedicines, while those obtained by *L. paracasei* using enzymatic-extracted extended-hydrolysis oligosaccharides were recognised as the leading antioxidants and anti-*H. pylori* phytomedicines. Incorporating these preparations into a barley malt beverage enhances antioxidant activity by approximately 20% while maintaining acceptable sensory characteristics. More broadly, this study supports the idea that specific plant substrates can be pharmacologically activated through defined microbial fermentation. It suggests that postbiotics generated from plant-derived materials serve as the active components of many plant-based therapeutics, offering a viable approach to creating standardised microbially matured phytomedicines from agro-industrial residues. Future steps toward clinical translation include in vivo pharmacokinetic studies, targeted metabolomics, and formulation optimisation. However, to fully realise their potential and ensure their effectiveness, additional research is needed to address the identified limitations.

Author Contributions: Conceptualisation, M.S. and S.H.R.; methodology, F.N., M.S., S.H.R. and M.M.; investigation and formal analysis, F.N.; resources, M.S., S.H.R., M.D.A.S., M.J.S., R.L. and N.M.D.; writing—original draft preparation, F.N.; writing—review and editing, M.S., S.H.R., M.M., M.D.A.S., M.J.S., R.L., M.C.M.III, S.Z. and N.M.D.; supervision, M.S., S.H.R., M.D.A.S., M.J.S., R.L. and N.M.D.; project administration, M.S.; funding acquisition, M.S. All authors have read and agreed to the published version of the manuscript.

Funding: This research was financially supported by a research grant from the University of Tehran. M.J.S., M.D.A.S., R.L. and N.M.D. acknowledge support from the University of Alberta.

Institutional Review Board Statement: The study was conducted in accordance with the Declaration of Helsinki and approved by the Research Ethics Committee of the University of Tehran (Approval No. ETHIC-202601-1665) on 1 November 2025.

Informed Consent Statement: Informed consent was obtained verbally from all participants involved in the study prior to their participation. Verbal consent was used in place of written consent, and this procedure was approved by the relevant research ethics committee.

Data Availability Statement: The original contributions presented in this study are included in the article. Further inquiries can be directed to the corresponding authors.

Acknowledgments: The authors thank Oghab Halva Company (Shahriar, Iran) for providing the sesame hull material, and the laboratory staff of the Functional Food Research Core, University

of Tehran, for technical assistance. During the preparation of this manuscript, the authors used a large language model (Claude 3.5, Anthropic) for editorial assistance with language, structure, and reference formatting. The authors have reviewed and edited the output and take full responsibility for the content of this publication.

Conflicts of Interest: The authors declare no conflicts of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results. Oghab Halva Company provided the product used in this study. The company had no role in the design of the study, the collection, analysis, or interpretation of the data, the writing of the manuscript, or the decision to publish the results.

References

1. World Health Organization. *WHO Guidelines on Good Herbal Processing Practices for Herbal Medicines*; WHO Technical Report Series, No. 1010, Annex 1; WHO: Geneva, Switzerland, 2018.
2. Matsumura, Y.; Kitabatake, M.; Kayano, S.I.; Ito, T. Dietary phenolic compounds: Their health benefits and association with the gut microbiota. *Antioxidants* **2023**, *12*, 880. [CrossRef]
3. Lippolis, T.; Cofano, M.; Caponio, G.R.; De Nunzio, V.; Notarnicola, M. Bioaccessibility and bioavailability of diet polyphenols and their modulation of gut microbiota. *Int. J. Mol. Sci.* **2023**, *24*, 3813. [CrossRef]
4. Williamson, G.; Clifford, M.N. Role of the small intestine, colon and microbiota in determining the metabolic fate of polyphenols. *Biochem. Pharmacol.* **2017**, *139*, 24–39. [CrossRef]
5. Martins, M.; Sganzerla, W.G.; Forster-Carneiro, T.; Goldbeck, R. Recent advances in xylo-oligosaccharides production and applications: A comprehensive review and bibliometric analysis. *Biocatal. Agric. Biotechnol.* **2023**, *47*, 102608. [CrossRef]
6. Salminen, S.; Collado, M.C.; Endo, A.; Hill, C.; Lebeer, S.; Quigley, E.M.M.; Sanders, M.E.; Shamir, R.; Swann, J.R.; Szajewska, H.; et al. The International Scientific Association of Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of postbiotics. *Nat. Rev. Gastroenterol. Hepatol.* **2021**, *18*, 649–667. [CrossRef]
7. Siciliano, R.A.; Reale, A.; Mazzeo, M.F.; Morandi, S.; Silvetti, T.; Brasca, M. Paraprobiotics: A new perspective for functional foods and nutraceuticals. *Nutrients* **2021**, *13*, 1225. [CrossRef]
8. Teame, T.; Wang, A.; Xie, M.; Zhang, Z.; Yang, Y.; Ding, Q.; Gao, C.; Olsen, R.E.; Ran, C.; Zhou, Z. Paraprobiotics and postbiotics of probiotic Lactobacilli: Effects on the host and action mechanisms. *Front. Nutr.* **2020**, *7*, 570344. [CrossRef]
9. De Simone, C. The unregulated probiotic market. *Clin. Gastroenterol. Hepatol.* **2019**, *17*, 809–817. [CrossRef]
10. Aggarwal, S.; Sabharwal, V.; Kaushik, P.; Joshi, A.; Aayushi, A.; Suri, M. Postbiotics: From emerging concept to application. *Front. Sustain. Food Syst.* **2022**, *6*, 887642. [CrossRef]
11. Barros, C.P.; Guimarães, J.T.; Esmerino, E.A.; Duarte, M.C.K.H.; Silva, M.C.; Silva, R.; Ferreira, B.M.; Sant'Ana, A.S.; Freitas, M.Q.; Cruz, A.G. Paraprobiotics and postbiotics: Concepts and potential applications in dairy products. *Curr. Opin. Food Sci.* **2020**, *32*, 1–8. [CrossRef]
12. Cuevas-González, P.F.; Liceaga, A.M.; Aguilar-Toalá, J.E. Postbiotics and paraprobiotics: From concepts to applications. *Food Res. Int.* **2020**, *136*, 109502. [CrossRef] [PubMed]
13. Vinderola, G.; Sanders, M.E.; Salminen, S. The concept of postbiotics. *Foods* **2022**, *11*, 1077. [CrossRef]
14. Zhao, Y.-S.; Eweys, A.S.; Zhang, J.-Y.; Zhu, Y.; Bai, J.; Darwesh, O.M.; Zhang, H.-B.; Xiao, X. Fermentation affects the antioxidant activity of plant-based food material through the release and production of bioactive components. *Antioxidants* **2021**, *10*, 2004. [CrossRef]
15. Espín, J.C.; González-Sarrías, A.; Tomás-Barberán, F.A. The gut microbiota: A key factor in the therapeutic effects of (poly)phenols. *Biochem. Pharmacol.* **2017**, *139*, 82–93. [CrossRef] [PubMed]
16. Possemiers, S.; Bolca, S.; Verstraete, W.; Heyerick, A. The intestinal microbiome: A separate organ inside the body with the metabolic potential to influence the bioactivity of botanicals. *Fitoterapia* **2011**, *82*, 53–66. [CrossRef]
17. Wegh, C.A.M.; Geerlings, S.Y.; Knol, J.; Roeselers, G.; Belzer, C. Postbiotics and their potential applications in early life nutrition and beyond. *Int. J. Mol. Sci.* **2019**, *20*, 4673. [CrossRef] [PubMed]
18. Li, H.-Y.; Zhou, D.-D.; Gan, R.-Y.; Huang, S.-Y.; Zhao, C.-N.; Shang, A.; Xu, X.-Y.; Li, H.-B. Effects and mechanisms of probiotics, prebiotics, synbiotics, and postbiotics on metabolic diseases targeting gut microbiota: A narrative review. *Nutrients* **2021**, *13*, 3211. [CrossRef]
19. Zhou, P.; Chen, C.; Patil, S.; Dong, S. Unveiling the therapeutic symphony of probiotics, prebiotics, and postbiotics in gut–immune harmony. *Front. Nutr.* **2024**, *11*, 1355542. [CrossRef]
20. Miran, M.; Salami, M.; Yarmand, M.S.; Ferreira-Lazarte, A.; Ariaeenejad, S.; Montilla, A.; Moreno, F.J. Arabinoxylo-oligosaccharide production from unexploited agro-industrial sesame (*Sesamum indicum* L.) hulls waste. *Carbohydr. Polym.* **2024**, *342*, 122399. [CrossRef]

21. Ávila, P.F.; Martins, M.; de Almeida Costa, F.A.; Goldbeck, R. Xylo-oligosaccharides production by commercial enzyme mixture from agricultural wastes and their prebiotic and antioxidant potential. *Bioact. Carbohydr. Diet. Fibre* **2020**, *24*, 100234. [CrossRef]
22. Palaniappan, A.; Antony, U.; Emmambux, M.N. Current status of xylo-oligosaccharides: Production, characterization, health benefits and food application. *Trends Food Sci. Technol.* **2021**, *111*, 506–519. [CrossRef]
23. Miran, M.; Salami, M.; Emam-Djomeh, Z.; Moreno, F.J.; Montilla, A. Isolation and structural evaluation of pectin, pectin-based polymer blends, composites, IPNs and gels. In *Handbook of Natural Polymers*; Sreekala, M.S., Ravindran, L., Goda, K., Thomas, S., Eds.; Elsevier Inc.: Amsterdam, The Netherlands, 2023; Volume 1, pp. 369–398. [CrossRef]
24. Álvarez, C.; González, S.; Ballesteros, I.; Gullón, B.; Negro, M. In vitro assessment of the prebiotic potential of xylo-oligosaccharides from barley straw. *Foods* **2023**, *12*, 83. [CrossRef]
25. Falck, P.; Linares-Pastén, J.A.; Karlsson, E.N.; Adlercreutz, P. Arabinoxylanase from glycoside hydrolase family 5 is a selective enzyme for production of specific arabinoxylo-oligosaccharides. *Food Chem.* **2018**, *242*, 579–584. [CrossRef]
26. Scheller, H.V.; Ulvskov, P. Hemicelluloses. *Annu. Rev. Plant Biol.* **2010**, *61*, 263–289. [CrossRef] [PubMed]
27. Lv, Q.-Q.; Cao, J.-J.; Liu, R.; Chen, H.-Q. Structural characterization, α-amylase and α-glucosidase inhibitory activities of polysaccharides from wheat bran. *Food Chem.* **2021**, *341*, 128218. [CrossRef]
28. Zheng, J.; Wittouck, S.; Salvetti, E.; Franz, C.M.A.P.; Harris, H.M.B.; Mattarelli, P.; O'Toole, P.W.; Pot, B.; Vandamme, P.; Walter, J.; et al. A taxonomic note on the genus *Lactobacillus*: Description of 23 novel genera, emended description of the genus *Lactobacillus* Beijerinck 1901, and union of *Lactobacillaceae* and *Leuconostocaceae*. *Int. J. Syst. Evol. Microbiol.* **2020**, *70*, 2782–2858. [CrossRef]
29. Abouloifa, H.; Khodaei, N.; Rokni, Y.; Karboune, S.; Brasca, M.; D'Hallewin, G.; Ben Salah, R.; Saalaoui, E.; Asehraou, A. The prebiotics (fructo-oligosaccharides and xylo-oligosaccharides) modulate the probiotic properties of *Lactiplantibacillus* and *Levilactobacillus* strains isolated from traditional fermented olive. *World J. Microbiol. Biotechnol.* **2020**, *36*, 185. [CrossRef] [PubMed]
30. Hsieh, F.-C.; Lan, C.-C.E.; Huang, T.-Y.; Chen, K.-W.; Chai, C.-Y.; Chen, W.-T.; Fang, A.-H.; Chen, Y.-H.; Wu, C.-S. Heat-killed and live *Lactobacillus reuteri* GMNL-263 exhibit similar effects on improving metabolic functions in high-fat diet-induced obese rats. *Food Funct.* **2016**, *7*, 2374–2388. [CrossRef]
31. Rozhkova, I.V.; Yurova, E.A.; Leonova, V.A. Evaluation of the amino acid composition and content of organic acids of complex postbiotic substances obtained on the basis of metabolites of probiotic bacteria *Lacticaseibacillus paracasei* ABK and *Lactobacillus helveticus* H9. *Fermentation* **2023**, *9*, 460. [CrossRef]
32. Munsch-Alatossava, P.; Rita, H.; Alatossava, T. A faster and more economical alternative to the standard plate count (SPC) method for microbiological analyses of raw milks. In *Communicating Current Research and Educational Topics and Trends in Applied Microbiology*; Méndez-Vilas, A., Ed.; FORMATEX: Badajoz, Spain, 2007; Volume 1, pp. 495–499.
33. Malunga, L.N.; Beta, T. Antioxidant capacity of arabinoxylan oligosaccharide fractions prepared from wheat aleurone using *Trichoderma viride* or *Neocallimastix patriciarum* xylanase. *Food Chem.* **2015**, *167*, 311–319. [CrossRef]
34. Gullón, B.; Eibes, G.; Moreira, M.T.; Dávila, I.; Labidi, J.; Gullón, P. Antioxidant and antimicrobial activities of extracts obtained from the refining of autohydrolysis liquors of vine shoots. *Ind. Crops Prod.* **2017**, *107*, 105–113. [CrossRef]
35. Spínola, V.; Llorent-Martínez, E.J.; Castilho, P.C. Inhibition of α-amylase, α-glucosidase and pancreatic lipase by phenolic compounds of *Rumex maderensis* (Madeira sorrel). *LWT* **2020**, *118*, 108727. [CrossRef]
36. Nabati, F.; Mojab, F.; Habibi-Rezaei, M.; Bagherzadeh, K.; Amanlou, M.; Yousefi, B. Large scale screening of commonly used Iranian traditional medicinal plants against urease activity. *DARU J. Pharm. Sci.* **2012**, *20*, 72. [CrossRef]
37. Nataraj, B.H.; Ali, S.A.; Behare, P.V.; Yadav, H. Postbiotics-parabiotics: The new horizons in microbial biotherapy and functional foods. *Microb. Cell Factories* **2020**, *19*, 168. [CrossRef]
38. Duarte, F.N.D.; Rodrigues, J.B.; da Costa Lima, M.; Lima, M.D.S.; Pacheco, M.T.B.; Pintado, M.M.E.; de Souza Aquino, J.; de Souza, E.L. Potential prebiotic properties of cashew apple (*Anacardium occidentale* L.) agro-industrial byproduct on *Lactobacillus* species. *J. Sci. Food Agric.* **2017**, *97*, 3712–3719. [CrossRef]
39. Schiraldi, C.; Valli, V.; Molinaro, A.; Cartenì, M.; De Rosa, M. Exopolysaccharides production in *Lactobacillus bulgaricus* and *Lactobacillus casei* exploiting microfiltration. *J. Ind. Microbiol. Biotechnol.* **2006**, *33*, 384–390. [CrossRef]
40. Miran, M.; Gashtasebi, S.; Salami, M.; Emam-Djomeh, Z.; Moreno, F.J. Biochemical properties and functions of pectin and hemicellulose polysaccharides and derived oligosaccharides: Impact on human health. In *Molecular Medicine and Biomedical Research in the Era of Precision Medicine*; Sobti, R.C., Wilson, D.W., Buttar, H.S., Telessy, I.G., Sobti, A., Eds.; Academic Press: London, UK, 2025; Volume 2, pp. 941–973. [CrossRef]
41. Freitas, C.D. Production of Xylo-Oligosaccharides by Different Treatments of Banana Pseudostem and Their Prebiotic Evaluation via Probiotic Bacteria Growth and Their Metabolites. Ph.D. Thesis, Programa de Pós-Graduação em Ciências Biológicas (Microbiologia Aplicada), Instituto de Biociências, Universidade Estadual Paulista (UNESP), São Paulo, Brazil, 2023.
42. Mladenov, M.; Lubomirov, L.; Grisk, O.; Avtanski, D.; Mitrokhin, V.; Sazdova, I.; Keremidarska-Markova, M.; Danailova, Y.; Nikolaev, G.; Konakchieva, R.; et al. Oxidative stress, reductive stress and antioxidants in vascular pathogenesis and aging. *Antioxidants* **2023**, *12*, 1126. [CrossRef] [PubMed]

43. You, S.; Ma, Y.; Yan, B.; Pei, W.; Wu, Q.; Ding, C.; Huang, C. The promotion mechanism of prebiotics for probiotics: A review. *Front. Nutr.* **2022**, *9*, 1000517. [CrossRef] [PubMed]
44. Bhoyar, M.S.; Mishra, G.P.; Naik, P.K.; Srivastava, R. Estimation of antioxidant activity and total phenolics among natural populations of caper (*Capparis spinosa*) leaves collected from cold arid desert of trans-Himalayas. *Aust. J. Crop Sci.* **2011**, *5*, 912–919.
45. Lasrado, L.D.; Gudipati, M. Antioxidant property of synbiotic combination of *Lactobacillus* sp. and wheat bran xylo-oligosaccharides. *J. Food Sci. Technol.* **2015**, *52*, 4551–4557. [CrossRef] [PubMed]
46. Choi, S.S.; Kim, Y.; Han, K.S.; You, S.; Oh, S.; Kim, S.H. Effects of *Lactobacillus* strains on cancer cell proliferation and oxidative stress in vitro. *Lett. Appl. Microbiol.* **2006**, *42*, 452–458. [CrossRef]
47. Li, S.; Pan, J.; Hu, X.; Zhang, Y.; Gong, D.; Zhang, G. Kaempferol inhibits the activity of pancreatic lipase and its synergistic effect with orlistat. *J. Funct. Foods* **2020**, *72*, 104041. [CrossRef]
48. Fan, Z.; Jia, W. *Lactobacillus casei*-derived postbiotics inhibited digestion of triglycerides, glycerophospholipids and sterol lipids via allosteric regulation of BSSL, PTL and PLA2 to prevent obesity: Perspectives on deep learning integrated multi-omics. *Food Funct.* **2023**, *14*, 7439–7456. [CrossRef]
49. Kittibunchakul, S.; Yuthaworawit, N.; Whanmek, K.; Suttisansanee, U.; Santivarangkna, C. Health beneficial properties of a novel plant-based probiotic drink produced by fermentation of brown rice milk with GABA-producing *Lactobacillus pentosus* isolated from Thai pickled weed. *J. Funct. Foods* **2021**, *86*, 104710. [CrossRef]
50. Chia, T.Y.; Gan, C.-Y.; Shafie, M.H.; Yap, P.G.; Rodhi, A.M.; Ahmad, A.; Murugaiyah, V.; Abdulla, M.H.; Johns, E.J. A comprehensive review on the pancreatic lipase inhibitory peptides: A future anti-obesity strategy. *Electron. J. Gen. Med.* **2023**, *20*, em470. [CrossRef]
51. Papoutsis, K.; Zhang, J.; Bowyer, M.C.; Brunton, N.; Gibney, E.R.; Lyng, J. Fruit, vegetables, and mushrooms for the preparation of extracts with α-amylase and α-glucosidase inhibition properties: A review. *Food Chem.* **2021**, *338*, 128119. [CrossRef] [PubMed]
52. Suwanangul, S.; Jaichakan, P.; Narkprasom, N.; Kraithong, S.; Narkprasom, K.; Sangsawad, P. Innovative insights for establishing a synbiotic relationship with *Bacillus coagulans*: Viability, bioactivity, and in vitro-simulated gastrointestinal digestion. *Foods* **2023**, *12*, 3692. [CrossRef] [PubMed]
53. Yılmaz, T.; Şimşek, Ö. Potential health benefits of ropy exopolysaccharides produced by *Lactobacillus plantarum*. *Molecules* **2020**, *25*, 3293. [CrossRef]
54. Zubaidah, E.; Suryawira, Y.M.; Saparianti, E. Comparative study on production of exopolysaccharide (EPS) by lactic acid bacteria (*L. casei* and *L. plantarum*) in different media (dates and mulberry juice). *Agroindustrial J.* **2014**, *3*, 107–115. [CrossRef]
55. Sun, L.; Zhao, H.; Liu, L.; Wu, X.; Gao, Q.; Zhao, Y. Effects of *Lactobacillus* on the inhibition of *Helicobacter pylori* growth. *Biotechnol. Biotechnol. Equip.* **2018**, *32*, 1533–1540. [CrossRef]
56. Duraisamy, S.; Sathyan, A.; Balakrishnan, S.; Subramani, P.; Prahalathan, C.; Kumarasamy, A. Bactericidal and non-cytotoxic activity of bacteriocin produced by *Lacticaseibacillus paracasei* F9-02 and evaluation of its tolerance to various physico-chemical conditions. *Environ. Microbiol.* **2023**, *25*, 2882–2896. [CrossRef] [PubMed]
57. Zacharof, M.-P.; Lovitt, R.W. Bacteriocins produced by lactic acid bacteria: A review article. *APCBEE Procedia* **2012**, *2*, 50–56. [CrossRef]
58. Hu, Y.; Zhu, Y.; Lu, N.-H. Novel and effective therapeutic regimens for *Helicobacter pylori* in an era of increasing antibiotic resistance. *Front. Cell. Infect. Microbiol.* **2017**, *7*, 168. [CrossRef] [PubMed]
59. Cian, R.E.; Garzón, A.G.; Albarracín, M.; Drago, S.R. Barley and malt as base ingredients for the production of new bio-functional foods. *Recent Prog. Nutr.* **2023**, *3*, 1. [CrossRef]
60. Akram, M.U.; Agunbiade, H.O.; Kadam, D.; Aluko, R.E.; Koksel, F. Adding Value to Brewery Industry By-Products as Novel Ingredients in Non-Alcoholic Malt Beverage Applications. *Foods* **2025**, *14*, 2882. [CrossRef]
61. Gupta, A.; Sanwal, N.; Bareen, M.A.; Barua, S.; Sharma, N.; Olatunji, O.J.; Nirmal, N.P.; Sahu, J.K. Trends in functional beverages: Functional ingredients, processing technologies, stability, health benefits, and consumer perspective. *Food Res. Int.* **2023**, *170*, 113046. [CrossRef] [PubMed]
62. Bolivar-Jacobo, N.A.; Reyes-Villagrana, R.A.; Arévalos-Sánchez, M.M.; Rentería-Monterrubio, A.L.; Santellano-Estrada, E.; Salas-Salazar, N.A.; Chávez-Martínez, A. Antioxidant and ACE-inhibition activities after in vitro digestion of a non-fermented dairy beverage enriched with postbiotics of *Lactobacillus* spp. *Fermentation* **2025**, *11*, 223. [CrossRef]

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