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Review

The role of the antimicrobial peptide nisin as a clean label food preservative

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The bacteriocin nisin can play a role in addressing the global need for safe, effective, and 'clean label' preservation strategies. Nisin A and its variants are among the most extensively studied antimicrobial peptides. Despite many advantages, nisin exhibits limitations in complex food matrices, including reduced solubility at neutral pH, susceptibility to proteolytic degradation, and poor activity against Gram-negative bacteria. We highlight recent advances aimed at overcoming these challenges, including novel delivery systems and the development of novel nisin variants with improved physicochemical properties, resistance to enzymatic degradation, and expanded antimicrobial spectra. Additionally, emerging research suggests a potential role for nisin as a functional food component capable of modulating the gut microbiome, although its effects appear context-dependent and require further investigation. We suggest that a diversified portfolio of nisin variants combined with advances in delivery strategies can position nisin and its variants as a key tool in the development of sustainable, safe, and minimally processed food.

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Introduction

Of the many bacteriocins that have been identified and characterized, nisin A and its variants are the most well studied. Nisin A is an amphiphilic, cationic lanthipeptide composed of 34 amino acids. Like all lanthipeptides, nisin contains post-translationally modified serine and threonine residues that are dehydrated to form dehydroalanine (Dha) and dehydrobutyrine (Dhb), respectively (Figure 2). Some of these modified residues react with neighboring cysteines to form lanthionine or methyllanthione rings that stabilize the peptide's structure and contribute to its antimicrobial activity [1]. Discovered by Rogers in 1928 [2], nisin was isolated and characterized as a peptide with potent antibacterial activity in the 1940s and developed for commercial use as a food preservative by the early 1950s [3].

Over time, regulatory agencies, including the WHO and USFDA, evaluated the safety of nisin and approved it for use as a food additive. It was the first bacteriocin to receive regulatory approval and is now used in over 80 countries [5]. Currently, nisin A and nisin Z are the only commercially available nisin variants used in food preservation. They are available as Nisaplin (Danisco, Dupont), Chrisin (Chr. Hansen), and vegetal or white NisinA/Z and NisinA/Z P (Handary). The global nisin market size was estimated at USD 502.9 million in 2023 and is projected to reach USD 691.1 million by 2030 [6]. As of now, nisin is widely used as a food preservative, with significant research efforts underway to expand its potential range of uses (Figure 1)

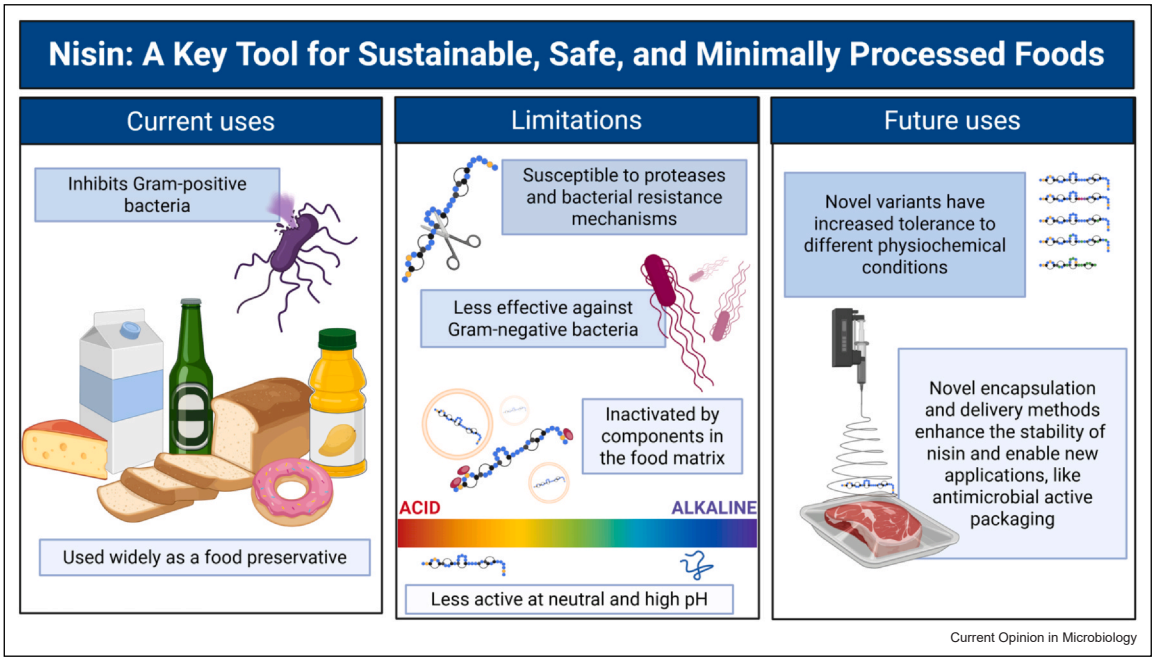
Nisin is a valuable tool in food production due to the broad range of foodborne pathogens it inhibits. Nisin acts by initially binding to lipid II, a component of the cell wall biosynthesis machinery. By segregating Lipid II, nisin interrupts the peptidoglycan production cycle and inhibits cell wall biosynthesis, in addition to forming pores in the bacterial membrane [7]. Nisin is significantly more active against Gram-positive bacteria. Activity against Gram-negative pathogens is restricted due to the presence of the LPS-containing outer membrane, which impedes the interaction between nisin and lipid II.

Current applications in food systems

Nisin is currently used in food systems as a targeted antimicrobial intervention, particularly where Gram-positive organisms and spore outgrowth are major risk factors [8]. It is also used to enable reductions in heat treatment intensity, salt concentrations, or other preservatives while maintaining safety.

A primary application is in dairy products, as nisin is compatible with dairy fermentation processes [9] and inhibits *Clostridium tyrobutyricum*, a spore-forming

Figure 1



The current and potential uses of the lantibiotic bacteriocin nisin. Created in BioRender. Hill, C. (2026) https://BioRender.com/naimey5.

bacterium responsible for late blowing defects in cheese that can result in significant economic losses. The addition of nisin prevents spore germination and improves product stability during storage [10].

In refrigerated ready-to-eat foods, nisin is commonly used as a post-lethality or surface-targeted control for *Listeria monocytogenes* [11]. Nisin is also applied in heat-processed and canned foods, where its ability to inhibit Gram-positive organisms and spore outgrowth can complement thermal processing [3], allowing manufacturers to use milder heat treatments for improved retention of nutritional and sensory quality [12].

Nisin as part of hurdle technology

In food preservation, hurdle technology is a method that integrates multiple preservation techniques to improve food safety, extend shelf life, and minimize the need for interventions that damage food sensory qualities. Nisin is frequently used as a component of hurdle technology, including physical treatments such as mild heating, UV irradiation, and ultrasound to control foodborne pathogens (Table 1). Nisin can be used in conjunction with heat treatment of foods to control foodborne pathogens such as *Listeria monocytogenes*. At pH 3, nisin can withstand temperatures used for High-Temperature, Short-Time pasteurization and autoclaving [13,14]. Nisin has also exhibited synergistic effects when combined with other bioactives, including antimicrobial peptides, plant extracts, antibiotics, antioxidants, chelating agents, biopolymers, and nanoparticles [15].

Limitations of nisin in food systems

Nisin has characteristics that limit its use in food systems (see Table 2). However, extensive research has been carried out to address these limitations, both by developing new ways to introduce nisin into food systems and by modifying the peptide itself. Current innovative approaches focus on expanding the functional range of nisin by enhancing attributes that make it particularly attractive for food applications, including solubility, pH tolerance, target specificity, antimicrobial activity, and stability.

Expanding the range of physiochemical conditions in which nisin is active

Food systems often possess physiochemical conditions that limit the effectiveness of nisin. While many studies demonstrate strong antimicrobial activity in lab settings, efficacy is frequently reduced in complex food matrices due to adsorption, partitioning, and degradation [39,49,50]. Nisin is most soluble at pH 3, with solubility decreasing as pH increases [3]. Structural modifications at key residues such as Dha5 can also significantly reduce antimicrobial activity [51].

To address these limitations, delivery systems have been developed to improve nisin performance in real food environments. Delivery technologies focus on ways to

Table 1

Recent advances in multi-hurdle technology used in combination with nisin.

Multi-hurdle technology	Activity	Usage
High hydrostatic pressure processing	*L. monocytogenes*	Controlling *L. monocytogenes* in oat milk [16]
Ultrasound, heat treatment	*B. subtilis* spores	Inactivating spores during sterilization [17]
Licochalcone A	*L. monocytogenes*	Controlling biofilm formation on food products [18]
Lecithin-nisin nanoparticles Chitosan nanoparticles Carvacrol nanoemulsions Lactoferrin-β-cyclodextrin hydrogels	*Escherichia coli, Salmonella enteritidis, Listeria monocytogenes, Staphylococcus aureus*, and *Campylobacter jejuni* biofilms	Food preservation and biofilm inhibition [19]
Zein nanofibers	*L. monocytogenes*	Active food packaging [20]
Cultured dextrose	*E. coli, S. aureus*	Preservative for meat systems [21]
Chinese chive seed extract	*S. aureus*	Controlling *S. aureus* in milk [22]
UV treatment	*Alicyclobacillus acidoterrestris* spores	Controlling beverage contaminants in orange juice and coconut water [23,24]
Lactoferrin-nisin nanoparticles	*L. monocytogenes, S. aureus, B. cereus*	Food preservation [25]
Nisin-curcumin nanoemulsion in starch-pectin film	*S. aureus, Salmonella*	Antimicrobial active salmon packaging [26]
Nisin-ZIF-8 composite integrated in polylactone/chitosan nanofibers	*E. coli, S. aureus*	Food packaging; extending fruit shelf life [27]
Xanthan gum and polyvinyl alcohol nanofibers	*S. aureus*	Active food packaging [28]
Bacterial cellulose films composited with alginate derivatives and nisin	*E. coli, S. aureus*	Biodegradable active food packaging [29]
Encapsulation of nisin Z by *L. lactis* L-WY410 extracellular vesicles	*L. monocytogenes, S. aureus*	Preservative within food matrices [30]
Oxidized cassava starch/ polycaprolactone/nisin ternary film	*S. aureus*	Biodegradable active food packaging [31]

protect nisin from degradation, improve its spatial distribution throughout food matrices, and enable controlled release to maintain effective concentrations over time. Encapsulation is an extensively studied approach, with recent research focusing on micro- and nanoscale carriers of nisin that enhance stability and activity within the food matrix and in packaging applications [52].

Encapsulation involves enclosing nisin within protective carriers such as liposomes, biopolymer particles, or nanocapsules. Materials like chitosan, alginate, and starch are commonly used due to their biodegradability and compatibility with food systems. These approaches have demonstrated clear benefits: nisin-loaded nanoparticles are more effective than free nisin at disrupting biofilms

Table 2

The limitations of nisin in food, and proposed solutions.

Challenge	Solution
Nisin is sensitive to digestion by proteinase K [3]	Cesin, a short natural nisin variant, is resistant to proteinase K digestion [32]
Nisin is sensitive to digestion by trypsin [3]	Salivaricin D is a trypsin-resistant bacteriocin similar to nisin [33] The nisin variant cesin R15G has increased resistance to trypsin [32]
Nisin's antilisterial activity can be reduced in the presence of high levels of fat and membrane components [34–36]	Encapsulation of nisin has the potential to protect nisin from inactivation by the food matrix, extending shelf life and active tie. For recent reviews of nisin encapsulation, refer to [37,38]
Nisin can be inactivated by emulsifiers and common additives in food such as sodium metabisulfate and titanium dioxide [3]	
Nisin is vulnerable to glycation and inactivation in the presence of sugar and heat [39]	
Nisin is less soluble and has lower activity at neutral and higher pH [3]	Nisin variants rombocin A, nisin Z and nisin F both have improved solubility at neutral pH compared to nisin A [40–42] Nisin P has optimal activity at neutral and basic pH [43]
Nisin is typically more effective against Gram-positive pathogens [3]	Outer membrane permeabilizing agents such as EDTA can improve nisin's activity against Gram-negative strains [44] Multiple novel and engineered variants have displayed increased activity against Gram-negative bacteria [9,45]
Nisin immunity systems such as the nisin resistance protein NSR can reduce nisin efficacy [46]	Nisin J has increased efficacy in inhibiting NSR-producing strains [47] Cesin is resistant to NSR due to its shortened structure [32]
Nisin has limited oxidative tolerance [3]	Nisin Q has improved oxidative tolerance [48]

[53], while lecithin-nisin nanoparticles more effectively prevented biofilm formation by five foodborne pathogens as compared to other nanoencapsulated bactericides [19].

Active packaging

Active packaging systems are those that are modified to extend shelf life and maintain or improve the condition of the food. Immobilization in a solid matrix can demonstrably improve the heat resistance and stability of nisin and provide extended antimicrobial protection in environments where free nisin would be inactivated [54,55].

Nanofiber-based systems are attractive due to their high loading capacity and adaptability [56]. Nisin-loaded nanofibers have demonstrated strong antimicrobial effects [56] and antioxidant activity [20], as well as the ability to extend the shelf life of perishable fruits [27,57]. Nisin can also improve the material qualities of nanofibers. The tensile strength of electrospun nanofibers composed of polyvinyl alcohol and oxidized xanthan gum fibers increased with higher nisin concentrations, a phenomenon attributed to Schiff base crosslinking [28]. Notably, one study observed that nisin-loaded PVA nanofibers inhibited *S. aureus* 1.5 years after manufacturing [56].

Conventional, non-biodegradable food packaging contributes to plastic waste pollution, leading to increased consumer interest in sustainable and biodegradable packaging solutions. A nisin-containing ternary composite polycaprolactone (PCL) film engineered with a solvent-free hot-pressing method actively inhibited *S. aureus* and displayed increased biodegradability as compared to pure PCL film [31].

Encapsulation of nisin within biodegradable films also enables use in raw meat systems, where free nisin would otherwise be inactivated. In raw meat, glutathione (GSH) can bind to the dehydroalanine residues in nisin, reducing antimicrobial activity [58,59]. However, nisin incorporated into electrospun nanofibers extended the shelf life of raw chicken [60], and nisin suspended in a biodegradable film with incorporated alginates was able to extend the shelf life of raw pork [29].

The delivery systems described above could expand nisin's role as a clean-label preservative by enhancing efficacy, enabling targeted and sustained activity, and reducing the need for excess use to offset food matrix limitations.

Nisin variants with improved characteristics

Bioengineering approaches have yielded novel nisin variants with enhanced activity, specificity, and stability across diverse conditions. Targeted amino acid substitutions can markedly modify antimicrobial activity and physicochemical properties, enabling the rational design of peptides suited to challenging food environments [8]. Naturally occurring nisin variants have also been discovered with enhanced physical properties relative to nisin A (for reviews, refer to Da Silva Oliveria et al. [61]).

Variants with improved stability at higher temperatures and broader pH ranges could extend the protective effects of nisin in food processing and storage environments. Natural variants nisin Z, nisin F, and nisin P already show improved solubility at neutral pH [41–43]. Bioengineered nisin variants have also displayed improved stability and solubility — including nisin A H27K and H31K [62] and nisin Z M21K and N20K [63], and nisin functionalized with a sugar moiety [64].

Other important targets for improvement include oxidative stability and production efficiency. Nisin Q retains higher levels of antimicrobial activity than nisin A under oxidizing conditions [48], suggesting that certain variants may be more suitable for oxidative environments. Increasing the acid-tolerance of *Lactococcus lactis* producer strains has been shown to improve nisin production yields [65].

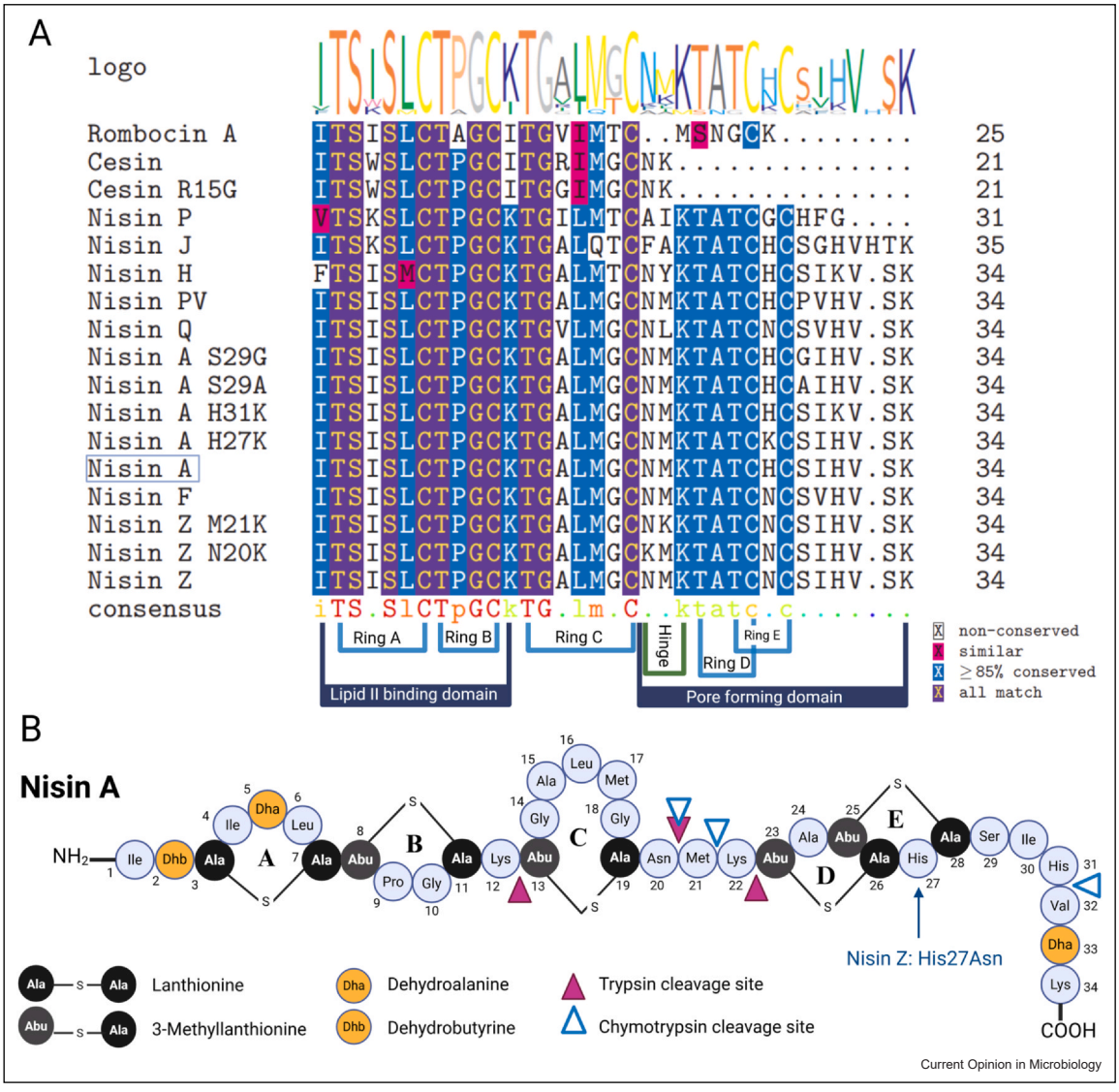
Increasing the tolerance of nisin to proteolytic degradation

Nisin is sensitive to proteolytic degradation by trypsin, chymotrypsin, and proteinase K, with several cleavage sites experimentally identified within the flexible hinge region (Figure 2) [3,4]. Simulated digestion studies using the INFOGEST protocol further demonstrated extensive degradation of both nisin A and nisin Z by gastrointestinal enzymes [66,67]. Enhancing resistance to enzymatic breakdown could prove an important strategy for improving the stability and effectiveness of nisin in food systems. One approach is to take advantage of variants with greater proteolytic stability. Salivaricin D is a 34-amino-acid nisin-like lantibiotic isolated from *Streptococcus salivarius* that is resistant to trypsin digestion [33]. Likewise, rombocin A is a natural nisin variant with anti-listerial properties that exhibits high resistance to proteolytic enzymes such as trypsin, chymotrypsin, and proteinase K [40]. Furthermore, Guo and colleagues were able to create modified nisin G18Dha that showed higher resistance to proteolytic enzymes compared to wild-type nisin [68].

Overcoming resistance mechanisms

One important area of investigation involves modifying nisin to overcome nisin-specific resistance mechanisms possessed by some spoilage and pathogenic bacteria [69]. The nisin resistance protein (NSR) proteolytically cleaves nisin, severely reducing its ability to form pores

Figure 2



Conserved and variable regions of natural and bioengineered nisin variants (a) Alignment of natural and bioengineered nisin variants discussed in the text. **(b)** Structure of the mature nisin A and nisin Z peptides, including trypsin and chymotrypsin cleavage sites determined experimentally by Slootweg et al. [4]. Post-translationally modified residues are shaded. Created in BioRender. Hill, C. (2026) https://BioRender.com/e2zsszs.

in the cell membrane [46]. Nisin resistance can develop in previously susceptible pathogens, underscoring the need for more robust nisin variants [70]. The natural variants nisin J [47] and cesin both display NSR resistance [32]. One bioengineered variant, nisin PV (S29P I30V), resists both NSR cleavage and oxidative inactivation, maintaining full antimicrobial activity against NSR-positive isolates [69].

Enhancing the activity of nisin against Gram-positive targets

Improving the efficacy of nisin against Gram-positive foodborne pathogens remains a key focus of bioengineering efforts. Nisin V (Nisin A M21V) and nisin A M21A both have single amino acid substitutions in the hinge region, which is critical for molecular flexibility and membrane insertion. Both variants exhibit enhanced activity against *L. monocytogenes* compared to nisin A in food model studies [71–73].

Combining bioengineered nisin variants represents another effective strategy to improve antimicrobial performance. Nyhan et al. reported derivatives (M17Q + N20P and M17Q + S29E) that exhibited enhanced anti-listerial activity in broth when used in combination than individually or compared to the nisin A producing strain [62].

Nisin H, produced by *Streptococcus hyointestinalis*, has increased activity against *Staphylococcus aureus*, illustrating the diversity of spectra in naturally occurring peptides with improved or altered antimicrobial profiles [74]. Indeed, it is anticipated that bioinformatic analysis of the rapidly expanding nisin family of peptides will provide clues about which parts of nisin are essential (conserved across variants) and which can be modified. A large genome mining study found variants with an alternative lipid II binding site and variants with a completely novel hinge region [75].

Increasing the activity of nisin against Gram-negative foodborne pathogens and spoilage organisms

Bioengineering can also produce variants that are more effective against Gram-negative strains. Nisin Z M21K and nisin Z N20K demonstrated improved antimicrobial activity against Gram-negative pathogens, *Shigella*, *Pseudomonas*, and *Salmonella*, possibly due to the presence of an extra positively charged lysine residue at the hinge region, which may enhance the interaction between nisin and the bacterial membrane [63]. Nisin A M21F and nisin A M17Q demonstrated enhanced antimicrobial activity against strains of the Gram-negative genus *Thermus* that cause pink discoloration in cheese [9]. Similarly, nisin A variants S29A and S29G have shown antimicrobial effects against a broader range of organisms, including *Cronobacter sakazakii*, *Escherichia coli* O157, and *Salmonella enterica*, in addition to Gram-positive targets like *Staphylococcus aureus*, possibly due to the enhanced ability of these variants to traverse the outer membrane relative to nisin A [45].

Nisin as a component of functional foods targeted at the microbiome

Functional foods are foods that can beneficially impact health and wellbeing or are implicated in disease risk reduction. Nisin has attracted growing interest as a potential functional food component due to its antimicrobial properties and its ability to influence microbial communities within the gastrointestinal tract (GIT). The newly described natural variant nisin G is being investigated as a probiotic due to its activity against *Fusobacterium nucleatum*, a Gram-negative pathogen associated with intestinal conditions [76].

Animal studies provide increasing evidence that dietary nisin can influence the structure and function of the gut microbiome. For example, orally ingested nisin A caused significant but reversible shifts in the gut microbiome of pigs [77]. In mice, oral administration of nisin A not only altered gut microbial composition but also influenced glucose metabolism [78] and norepinephrine expression in the brain [79]. Similarly, dietary supplementation with nisin A in rabbits produced dose-dependent changes in the microbiome [80]. Although nisin A and nisin Z are generally susceptible to degradation in the GIT due to pH conditions and proteolytic enzymes, high doses of nisin were shown to transit the pig GIT intact to at least some degree [77]. Delivery systems can also influence outcomes: when incorporated into starch-based matrices, nisin A was able to survive passage through the upper GIT and subsequently modify the microbiome of the lower gut in mice [81].

However, the effects of nisin on the microbiome are not universally beneficial and may depend on context and whether introduced as a purified product or in the form of a nisin-producing strain. For instance, in hyperglycaemic zebrafish models, treatment with Nisin A altered gut microbiome composition but also disrupted intestinal barrier integrity and triggered systemic inflammation [82].

Together, these findings suggest that nisin has potential as a functional food ingredient capable of modulating microbial communities in the GIT. However, further research is required to better understand its impact on, and ability to shape microbial communities, optimal delivery strategies, and long-term effects on host health.

Concluding remarks

Nisin is no longer a single molecule with a handful of close relatives. Recent genome mining studies have shown that nisin biosynthetic gene clusters are widespread across the biosphere, suggesting that a substantial and largely untapped reservoir of novel nisin variants is still waiting to be discovered [75]. Advances in genome-mining pipelines have already led to the discovery of novel nisin variants, including Lan-DF and Nisin VP [75,83].

Indeed, the growing availability of genomic data is facilitating a shift toward data-driven, rational design of nisin derivatives. A key finding is the apparent variability surrounding functional modules such as the hinge region, suggesting that structural features involved in membrane insertion and pore formation are evolving naturally while maintaining activity. This data will be invaluable for engineering objectives that aim to improve stability across a range of pH, salt, and temperature conditions, enhance solubility and diffusion in complex food matrices, and increase antimicrobial potency at low concentrations.

As more novel variants are discovered, a portfolio approach becomes conceivable. Rather than relying on nisin A and nisin Z alone, tailored combinations of nisin variants and delivery systems can be deployed for specific applications, potentially reducing selection pressure on individual peptides. In parallel, development of cost-

effective production systems will be critical to enable large-scale application of new variants, alongside research into the long-term impacts on microbial ecosystems.

Furthermore, greater integration with hurdle technologies and advanced delivery systems that use novel encapsulation strategies will ultimately boost the functional performance of nisin and strengthen its position as a trusted antimicrobial preservative. When addressing global food security, nisin has a place both in the food matrix and in active antimicrobial packaging systems.

Overall, the discovery and development of new nisin variants parallels antibiotic discovery, but with more targeted activity, fewer resistance mechanisms, and comparatively lower production costs — highlighting significant untapped potential for both food and agricultural applications.

Data Availability

No data were used for the research described in the article.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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- •• of outstanding interest

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