Review Article

Use of Enterocins-Producing Lactic Acid Bacteria for Food Biopreservation: Diversity and Functional Insights

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Abstract | Food safety and shelf-life stability continue to drive the search for natural antimicrobial agents for effective food biopreservation. Enterocins; a group of bacteriocins produced by *Enterococcus* species within lactic acid bacteria, exhibit high stability during food processing and strong inhibitory activity against major foodborne pathogens, particularly Gram-positive bacteria. Their antimicrobial action involves membrane interaction, receptor-mediated binding, pore formation, and disruption of cellular homeostasis. This review aims to integrate current knowledge on enterocin producer diversity and gene-cluster organization with their mechanisms of action, application formats, including protective cultures, purified and semi-purified peptides, emerging delivery systems, and associated safety and regulatory considerations. By linking molecular characteristics to practical applications and compliance requirements, this review provides a framework for the realistic implementation of enterocins as food biopreservatives.

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Introduction

Foodborne diseases and microbial spoilage remain major challenges in global food systems, threatening public health and causing substantial economic losses (Anumudu *et al.*, 2022). Concerns regarding long-term exposure to certain synthetic preservatives have increased interest in biopreservation, defined as the use of protective microorganisms and/or their metabolites to improve food safety and shelf life. In this context, bacteriocins produced by lactic acid bacteria (LAB) are widely studied natural antimicrobials that align with clean-label trends and industry needs (Putri *et al.*, 2024). Many LAB used as starter cultures and probiotics can produce bacteriocins, ribosomally synthesized antimicrobial peptides that are active at low concentrations, often heat stable, and generally degradable by digestive enzymes, supporting their suitability for food biopreservation (Ayivi *et al.*, 2020; Yap *et al.*, 2022).

Among LAB-derived bacteriocins, enterocins produced by the genus *Enterococcus* have attracted particular attention. *Enterococcus* species are frequently

found in fermented foods such as cheese, fermented sausages, and other traditional products. In these matrices, *Enterococcus* may contribute to flavor and texture development and produce enterocins with inhibitory activity against major foodborne pathogens, particularly *Listeria monocytogenes* and *Staphylococcus aureus* (Bhattacharya *et al.*, 2022). Inhibition of certain Gram-negative bacteria has also been reported, although this activity is typically observed under specific conditions, such as when the outer membrane becomes destabilized, in combination with other hurdles, or within complex food matrices rather than in pure culture systems. Kasimin *et al.* (2022) classified enterocins into four major groups (Classes I–IV), with most food-associated enterocins belonging to Class II; small, heat-stable, non-antibiotic peptides that retain activity across a broad range of pH values and food-processing conditions. This diversity provides multiple functional options that can be selectively applied to different food biopreservation contexts.

The potential of enterocin-producing LAB has been demonstrated both in laboratory settings and in real food systems. *Enterococcus faecium* strains have been shown to reduce *L. monocytogenes* counts in meat and milk, suppress spoilage microorganisms, and extend food shelf life without adversely affecting sensory attributes (Hernández-González *et al.*, 2021; Bhattacharya *et al.*, 2022). A recent study reported that *E. faecium* incorporated into alginate films, together with enterocins, has effectively reduced *Salmonella enterica* populations in chicken meat (Rashid *et al.*, 2023). These findings reinforce the idea that enterocins may complement or even partially replace chemical preservatives in high-risk animal-derived foods (Putri *et al.*, 2024).

Despite these advantages, *Enterococcus* is a "dual-nature" genus. While being beneficial in fermentation, flavor development, and has potential probiotic attributes, some strains are known opportunistic pathogens that may harbor virulence genes and antibiotic resistance traits, including vancomycin-resistant Enterococci (VRE). Yordanova *et al.* (2024) emphasized that any enterocin-producing *Enterococcus* strain intended for food applications must undergo rigorous safety evaluation, including antibiotic susceptibility profiling, virulence gene screening, and genomic characterization to clearly distinguish safe strains from clinically concerning lineages. This balance between high antimicrobial potential and safety considerations underscores the scientific and practical importance of enterocin research, demanding careful alignment of antimicrobial efficacy, consumer safety, and regulatory compliance.

Given these complexities, an updated synthesis is needed to evaluate enterocin-producing LAB as a unified functional group rather than as isolated case studies. Unlike previous enterocin-focused reviews, this article integrates the taxonomic, ecological and genomic diversity of entercocin-producer strains, structural variation, gene-cluster organization, mechanisms of action in relation to antimicrobial spectrum and technological stability, and implementation pathways in real food systems (*i.e.*, protective cultures, purified/semi-purified enterocins, active packaging, and multi-hurdle preservation) together with explicit safety and regulatory considerations. By integrating these elements, this review aims to provide a holistic, practical-oriented understanding of enterocin biopreservation, and outlines strategic directions for safer, more sustainable application in modern food systems.

Diversity of enterocin-producing lactic acid bacteria

Genus- and species-level diversity of enterocin-producing lactic acid bacteria

Enterococci belonging to the genus *Enterococcus*, comprise several important species, particularly *E. faecalis* and *E. faecium*. These two species have been extensively studied due to their ability to produce a wide range of bacteriocins collectively known as enterocins. Numerous studies have reported that enterocin-producing enterococci are widely distributed across diverse environments, including the human gastrointestinal tract, fermented foods, and animal-derived products (Franz *et al.*, 2007; Javed *et al.*, 2011). For example, several *E. faecium* strains have been successfully isolated from various food sources such as dairy and meat products, highlighting their ecological significance within food-related ecosystems (Merzoug *et al.*, 2025). Previous studies reported by Birri *et al.* (2010), Settanni *et al.* (2014), Gupta *et al.* (2016), Abengozar *et al.* (2017), Al-Madboy *et al.* (2020), García-Vela *et al.* (2024) and Souza *et al.* (2024) demonstrated that some enterocins exhibit broad-spectrum antimicrobial activity, whereas others act more narrowly, indicating functional specialization associated with their ecological roles, as summarized in Table 1.

Table 1: *Enterocin producing lactic acid bacteria: Diversity and functional characteristics.*

Genus / species	Enterocin type	Antimicrobial spectrum	Special characteristics	References
Enterococcus faecium	Enterocin A, B, P, L50A/B	*Listeria monocytogenes, Streptococcus suis, Streptococcus pyogenes, Enterococcus cecorum, Enterococcus faecalis, Campylobacter coli, Pseudomonas aeruginosa*	Stable under heat and pH variations, active across a broad spectrum.	Garcia-Vela *et al.* (2024)
Enterococcus faecalis	Enterocin AS-48	*Listeria monocytogenes, Clostridium perfringens, Leishmania donovani*	Unique cyclic structure (AS-48), high stability, effective at low concentrations.	Abengozar *et al.* (2017)
Enterococcus durans MF5	Enterocin MF5	*Listeria monocytogenes, Listeria innocua, Listeria seeligeri, Listeria grayi, Listeria ivanovii*	Heat-resistant; stable across a wide pH range; active against *Listeria* spp. by disrupting bacterial cell membranes	Souza *et al.* (2024)
Enterococcus hirae LD3	Enterocin LD3	*Listeria monocytogenes, Salmonella enterica, Staphylococcus aureus, Pseudomonas fluorescens, Pseudomonas aeruginosa, Salmonella typhi, Shigella flexneri, E. coli O157:H7, Vibrio* spp.	Broad antimicrobial spectrum, active against both Gram-positive and Gram-negative bacteria, activity unaffected by proteolytic enzymes (papain, proteinase K, pepsin, trypsin).	Gupta *et al.* (2016)
Enterococcus mundtii WFE3, WFE20, WFE31	Mundticin KS	*Listeria monocytogenes*	Stable across a broad pH range, stable in ethanol; non-adsorptive to producer cell surface, bactericidal effect; optimal production at neutral pH and 30–37 °C; non-cytotoxic, producer strain remains sensitive to common antibiotics.	Settanni *et al.* (2014)
Enterococcus thailandicus	Enterocin LNS18	*Active against various pathogenic bacteria (broad spectrum)*	Exhibits broad-spectrum antimicrobial activity and potential anticancer properties.	Al-Madboly *et al.* (2020)
Enterococcus avium	Avicin A	*Listeria* spp., *Listeria monocytogenes*	Strong antimicrobial activity, structural similarity to Mundticin KS and Enterocin CRL35.	Birri *et al.* (2010)

Genetic and biochemical characterization of enterocins reveals a complex classification system primarily based on their structural and functional properties. Class IIa bacteriocins, for example, are well known for their strong activity against various Gram-positive pathogens and play a crucial role in food preservation and safety (Ennahar *et al.*, 2000). Enterocin production by different *Enterococcus* species has been widely documented to inhibit pathogens such as *L. monocytogenes* and *Staphylococcus aureus*, reinforcing the potential of enterocins as effective biopreservative agents in food technology (Herranz and Driessen, 2005; Jaouani *et al.*, 2014). A thorough understanding of the genetic basis underlying enterocin production is essential, as it provides valuable insights into the biosynthetic pathways and regulatory mechanisms governing their secretion (Park *et al.*, 2003).

Enterocin-producing *Enterococcus* strains display remarkable adaptability across diverse habitats, ranging from food matrices and gastrointestinal tracts of animals to the human microbiota. The functional diversity of enterocins enables these *Enterococcus* strains to survive in competitive environments where they must inhibit or evade antimicrobial actions from other microorganisms. For instance, bacteriocins produced by *E. faecium* have been shown to suppress the growth of other enterococci, including antibiotic-resistant strains, contributing to the structure and dynamics of the microbial communities (Izquierdo *et al.*, 2009). Additionally, environmental factors such as pH and temperature are known to influence bacteriocin production and activity, ultimately affecting the bacterial ecological competitiveness and survival (Costa *et al.*, 2019; Alang *et al.*, 2020).

Structural and functional classification of enterocins

Bacteriocins produced by enterococci are classified into several structural groups and share many similarities with bacteriocin classes produced by other LAB, as summarized in Table 2. Traditionally, bacteriocins are grouped into three major classes. Class I includes cyclic bacteriocins, with the classic example being enterocin AS-48; a 70–amino acid cyclic peptide produced by *E. faecalis*. This enterocin exhibits exceptional stability against heat and a wide range of pH conditions, and demonstrates broad-spectrum activity against Gram-positive bacteria such as *L. monocytogenes* and *Staphylococcus aureus* (Kurushima *et al.*, 2014).

The cyclic structure of enterocin AS-48 contributes appreciably to its stability, making it highly relevant for preventing microbiological spoilage of foods (Martínez-Bueno *et al.*, 1994; Vimont *et al.*, 2017).

Class II bacteriocins are smaller peptides and are further divided into several subclasses based on their structural characteristics and target specificity. Class IIa bacteriocins including pediocin like peptides, such as enterocin A, P, and 96 are characterized by a conserved N terminal "YGNGV" motif. These peptides possess strong anti-Listerial activity, particularly important for food safety applications (Birri *et al.*, 2010). Production of these bacteriocins reflects the evolutionary advantage of enterococci in competing with other microorganisms in their environment (Tuncer *et al.*, 2013).

Class IId consists of single-peptide bacteriocins that lack a leader sequence (leaderless), represented by enterocin Q and enterocin DD14. These peptides are synthesized without a precursor leader and express unique properties, such as receptor-independent activity and membrane-targeting mechanisms that collectively enhance their antimicrobial effectiveness (Ladjouzi *et al.*, 2020). This subclass highlights the versatility of enterococci in producing a wide array of antimicrobial agents, enabling them to adapt and thrive across diverse ecological niches (Liu *et al.*, 2011).

Table 2: *Representative enterocins: Structural classes, producing species, and key functional features.*

Enterocin	Structural class/ type	Typical producing species	Main targets / antimicrobial spectrum	Notable properties and applications	Selected references
AS-48	Class I, circular bacteriocin	*E. faecalis*	Broad Gram-positive spectrum, including Listeria and Staphylococcus	Exhibits exceptional heat and pH stability, extensively studied in dairy and meat systems, promising for milk and cheese biopreservation.	Franz *et al.* (2007)
Enterocin A	Class IIa, pediocin-like	*E. faecium*	Strong anti-Listeria activity; active against other Gram-positive bacteria	Contains the YGNGV motif, often co-produced with other enterocins, suitable candidate for synthetic heterologous production.	Wu *et al.* (2022)
Enterocin P	Class IIa, pediocin-like	*E. faecium* (e.g., strain L50)	Listeria spp., some Bacillus and Staphylococcus	~4.6 kDa peptide, chromosomally encoded in strain L50, active in diverse food matrices.	Ness *et al.* (2014)
Enterocin 96	Class II, pediocin-like bacteriocin	*E. faecalis*	Gram-positive pathogens, including Listeria	Demonstrated efficacy, well-characterized structure and activity profiles.	Izquierdo *et al.* (2009)
L50A/B	Class IId, leaderless, two-peptide	*E. faecium L50*	Broad Gram-positive spectrum; potent activity against Listeria	Synthesized without a leader peptide, encoded by plasmid, receptor-independent mechanism, model leaderless bacteriocin.	Ness *et al.* (2014)
Enterocin Q	Class IId, leaderless	*E. faecium L50*	Gram-positive bacteria including Listeria spp.	Encoded on plasmid pCIZ2, frequently co-produced with L50 and P in multi-enterocin strains.	Ness *et al.* (2014)
EJ97	Class IId, leaderless	*E. faecalis EJ97*	Listeria monocytogenes and related species	Well-documented anti-Listeria enterocin, representative of leaderless single-peptide bacteriocins.	Wu *et al.* (2022)
DD14	Class IId, leaderless, two-peptide	*E. faecalis*	Clostridium perfringens and other Gram-positive bacteria	Considered a "safe" leaderless enterocin, recommended for anti-Clostridium use and food bioprotection applications.	Luenglusontigit *et al.* (2023)
ESmr18 (Enterocin Smr18)	Class II-like bacteriocin	*E. faecium Smr18*	Salmonella enterica and Gram-positive bacteria	Semi-purified form shown effective in chicken meat, supports enterocin application against Gram-negative bacteria under certain conditions.	Rashid *et al.* (2023)
Multiple enterocins (A, B, P, L50-like)	Mixed IIa + leaderless	*E. faecium* from dairy and artisanal foods	Broad Gram-positive spectrum; frequently strong anti-Listeria activity	Multi-enterocin-producing strains are common in raw milk and cheese, promising for food safety but require strict safety evaluation.	Merzoug *et al.* (2025)

Another subclass within Class II is Class IIb, comprising two-peptide leaderless enterocins. Enterocin L50A/B and MR10A/B are notable examples that function through synergistic interactions between peptide pairs, resulting in enhanced antimicrobial activity against target pathogens (Kurushima *et al*., 2014; Hanchi *et al*., 2016). This dual-peptide mechanism increases antibacterial potency by enriching the strategies by which enterocins disrupt target cells (Javed *et al*., 2010; Liu *et al*., 2011).

Understanding the diversity of these bacteriocins is crucial in microbiology and food technology, as it underscores the remarkable role of enterococci as beneficial microbial agents in food biopreservation and probiotic applications. The ability of enterococci to produce bacteriocins with substantial structural variability and strong functional potential presents promising opportunities for controlling foodborne pathogens. This multifunctionality positions enterococci as compelling candidates for further research in both pharmaceutical and food industries, and emphasizes the need for continued exploration of the biochemical pathways and genetic frameworks that regulate bacteriocins production.

Enterocin gene clusters

Enterocin gene clusters are a fundamental component of the antimicrobial capacity of enterococci, particularly *E. faecium* and *E. faecalis*, well known for producing multiple bacteriocins. These clusters typically contain structural genes encoding prepeptides, immunity determinants, and additional genes involved in modification and transport. Structural genes such as *entA*, *entB*, *entL50A/B*, and *entQ* encode precursors of diverse enterocins produced by different *Enterococcus* strains (Toğay *et al*., 2016; Liu *et al*., 2019). Organization of these genes into operon-like clusters enables coordinated expression of production, secretion, and self-immunity functions, essential for efficient bacteriocin biosynthesis, and represents a key determinant of functional output in food systems.

Genomic analyses have shown that many enterocin gene clusters are located on plasmids, a feature that has important implications for both biosafety assessment and industrial application. Plasmid localization facilitates horizontal gene transfer among enterococcal strains, enhancing ecological adaptability but simultaneously increasing the risk of co-transfer of undesirable traits such as antimicrobial-resistance or virulence determinants. From a food-biopreservation perspective, this genetic context necessitates genome-level screening to distinguish strains suitable for direct use as starter or protective cultures from those better suited for controlled fermentation or purified enterocin production. For example, *E. faecium* L50 harbors plasmid-encoded genes for enterocins L50 and Q_2 alongside chromosomal genes encoding enterocin P (Belgacem *et al*., 2010), illustrating how mixed genomic localization can complicate biosafety evaluation.

Multiple enterocin gene clusters may co-exist within a single strain, particularly in isolates from raw milk and artisanal dairy products, potentially broadening antimicrobial spectra (Liu *et al*., 2019; Lopes *et al*., 2024). However, the industrial feasibility of exploiting such strains depends not only on gene presence but also on regulatory stability and absence of mobile genetic elements linked to clinically relevant traits. Immunity genes, typically located adjacent to their corresponding structural genes, protect producer strains from autotoxicity and further underscore the importance of cluster integrity for predictable and safe bacteriocin expression (Cintas *et al*., 2000; Sanchez *et al*., 2008).

Although enterocin genes are widely distributed, but their presence does not always correlate with detectable antimicrobial activity, as expression may be modulated by mutations, regulatory elements, or environmental conditions. Consequently, strain selection for food or biotechnological applications must integrate genomic architecture with phenotypic validation. Variability in enterocin gene profiles across the strains suggests that specific gene-cluster combinations may enhance adaptive fitness, y*et al*so requires careful evaluation to balance antimicrobial efficacy with biosafety considerations (Hill *et al*., 2020; Lopes *et al*., 2024).

The evolutionary dynamics of enterocin gene clusters further reinforce their relevance to biosafety and industrial deployment. Horizontal gene transfer promotes rapid dissemination of bacteriocin genes under selective pressure, contributing to genetic diversity in competitive ecosystems such as fermented foods and animal-derived products (Hill *et al*., 2020; Lopes *et al*., 2024). While this plasticity supports antimicrobial activity, it also underscores the need to select genetically stable production platforms,

particularly when enterocins are intended for large-scale food preservation or therapeutic use (Belguesmia *et al*., 2010; Oladipo *et al*., 2015). Overall, enterocin gene clusters represent adaptive genomic elements whose organization directly influences antimicrobial performance, biosafety risk, and the feasibility of industrial application.

Functional insights: Mechanisms and biological activities

Antimicrobial mechanisms of enterocins against pathogenic bacteria

Enterocins exert antimicrobial activity primarily through targeted interactions with the bacterial cell membrane, with mechanistic features that directly influence their performance in food systems. Initial electrostatic attraction between the cationic peptide and negatively charged membrane components facilitates binding to specific receptors, most notably the mannose phosphotransferase system (Man-PTS), which is prevalent in Gram-positive pathogens such as *L. monocytogenes* and *Staphylococcus aureus* (Alvarez-Sieiro *et al*., 2016; Chikindas *et al*., 2018). This receptor-dependent mechanism explains the consistently strong activity of Class IIa enterocins against *Listeria* spp.; a key target in ready-to-eat and minimally processed foods, and also accounts for their limited efficacy against Gram-negative bacteria due to the protective outer lipopolysaccharide layer (Hanchi *et al*., 2018).

Following receptor engagement, enterocins become insert into the cytoplasmic membrane and form pores that disrupt membrane integrity. Class IIa enterocins typically form small, receptor-guided pores, whereas Class IIb enterocins require the synergistic action of two peptides to generate larger and more stable pore complexes (Darbandi *et al*., 2022). These mechanistic differences influence antimicrobial potency, spectrum, and dose requirements in complex food matrices. Pore formation leads to leakage of ions and metabolites, loss of membrane potential, and rapid collapse of cellular homeostasis (Yap *et al*., 2022; Wang *et al*., 2023), resulting in bactericidal effects without extensive cell lysis; a feature advantageous for maintaining food quality.

Beyond membrane disruption, enterocins efficacy in food systems is enhanced by their physicochemical stability. Many enterocins retain activity across a broad pH range, tolerate high temperatures, and remain functional under common food-processing and storage conditions, including pasteurization and refrigeration (Abanoz and Kunduhoglu, 2018; Kasimin *et al*., 2022). Collectively, receptor specificity, pore-forming strategy, and environmental stability represent the key mechanistic determinants governing enterocins performance as natural biopreservatives. A comprehensive understanding of these processes is illustrated in Figure 1.

Antimicrobial spectrum and technological robustness

Enterocins are known for their strong inhibitory activity against a range of Gram-positive pathogens, particularly *L. monocytogenes*, *Staphylococcus aureus*, and other coagulase-negative staphylococci (Turhan *et al*., 2020; Souza *et al*., 2024). This broad antimicrobial spectrum is partly attributed to the diverse modes of action exhibited by enterocins, including pore formation in bacterial membranes and disruption of cellular homeostasis, both of which are key factors underlying their effectiveness against Gram-positive and, to a lesser extent, Gram-negative bacteria (Garmasheva and Oleschenko, 2023).

Advances in food processing technologies have further strengthened the potential of enterocins as promising biopreservatives. In general, enterocins have demonstrated substantial heat stability, retaining antimicrobial activity even after exposure to temperatures between 60–100°C for short durations (Zommiti *et al*., 2018; Souza *et al*., 2024). This heat resistance is particularly valuable for food applications involving thermal treatments. In addition, enterocins remain active across a broad pH range (approximately pH 3–8), providing flexibility for use in various food matrices and environmental conditions (Belguesmia *et al*., 2010; Souza *et al*., 2024). Their pH stability supports widespread application in diverse food products and aligns well with consumer's preferences for natural and clean-label preservatives. Enterocins are also degraded by the gastrointestinal proteases, reducing the risk of accumulation or toxicity in the host after consumption (Kasimin *et al*., 2022; Souza *et al*., 2024).

Particular attention has been given to enterocins produced by *Enterococcus* strains capable of synthesizing multiple variants, as the combination of different enterocins may enhance overall antimicrobial activity. For example, *E. faecium* strains that simultaneously produce enterocins A and B display considerably increased activity against *Listeria*, demonstrating a

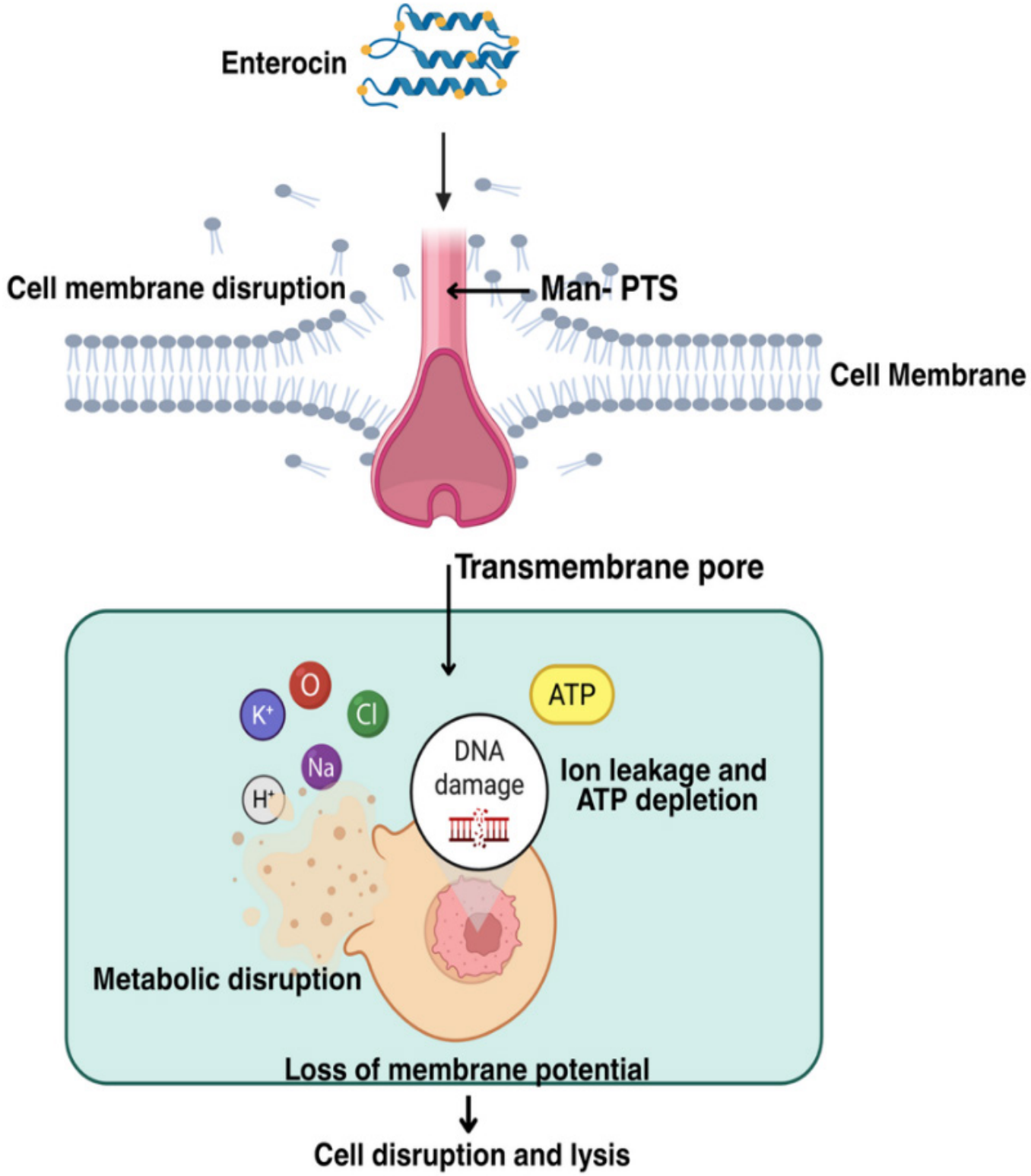

Figure 1: *Proposed antimicrobial mechanism of enterocins on bacterial cell membranes, adopted by Wang et al. (2024).*

clear synergistic effect (Mareková *et al.*, 2003; Turhan *et al.*, 2020). Findings related to such synergistic interactions open new opportunities for developing advanced biopreservation strategies, including novel formulations designed to maximize the antimicrobial potential of bacteriocins.

The strong efficacy of enterocins against a variety of pathogenic bacteria, along with their stability under diverse processing conditions, positions them as attractive natural alternatives to synthetic preservatives. Ongoing research into their mechanisms of action, synergistic applications, and technological enhancement will likely broaden their use across both the food and health sectors.

Additional functional traits of enterocin-producing strains

The functional traits of enterocin-producing *Enterococcus* strains contribute to their technological relevance in food fermentation, influencing both product stability and sensory development. Among these traits, rapid acidification and salt tolerance are particularly relevant, as they support performance in diverse food matrices, including dairy and fermented meat products. Previous studies have shown that *Enterococcus* spp. can tolerate high-salt environments commonly encountered in fermented meats, facilitating their persistence and metabolic activity during processing (Agüero *et al.*, 2020). Rapid acidification may contribute to shelf-life extension and improve microbial stability by creating conditions less favorable for spoilage and pathogenic microorganisms (Cirat *et al.*, 2024). However, these technological properties are strain-dependent and should not be generalized across the genus. Consequently, application of enterocin-producing *Enterococcus* strains in food systems requires careful strain-level characterization, with particular attention to the absence of clinically relevant virulence factors and transferable antimicrobial-resistance determinants.

In addition, *Enterococcus* strains can produce flavor-active metabolites and exopolysaccharides (EPS), playing important roles in shaping the texture and sensory quality of fermented foods. EPS production, for instance, has been associated with improved mouthfeel and viscosity in dairy products and other food matrices, ultimately affecting consumers acceptance (Bansal *et al.*, 2022; Jin *et al.*, 2025). Aromatic compounds generated during fermentation further contribute to desirable flavor profiles, enhancing the overall sensory appeal of the final product (Zhadyra *et al.*, 2025). These functional metabolites have demonstrated the potentials of enterococci in developing novel food formulations aimed at improving taste and texture while preserving health-promoting attributes.

Certain *Enterococcus* strains have also displayed the ability to survive under gastrointestinal (GI) conditions; a crucial prerequisite for probiotic applications. Their tolerance to gastric acidity and bile salts indicates their potential to exert beneficial health effects upon consumption (Nachér-Vázquez *et al.*, 2017). Additionally, several *Enterococcus* strains have shown to possess immunomodulatory properties, expressing that they can positively influence host immune responses and offer protective effects against gastrointestinal pathogens (Prajapati *et al.*, 2023). These findings underscore the dual functional roles of *Enterococcus* strains in food technology, as they not only enhance the quality and safety of food products but also offer potential health benefits when incorporated into the diet.

Applications of enterocin-producing LAB and enterocins in food biopreservation

Protective LAB cultures in fermented and minimally processed foods

The application of enterocin-producing lactic acid bacteria (LAB) as protective cultures has attracted growing interest as a natural strategy to enhance safety and extend shelf life in fermented and minimally processed foods. These strains exert antimicrobial effects through bacteriocin production, contributing to pathogen control in meat, poultry, and dairy systems while preserving product quality. Studies on meat and dairy matrices consistently have demonstrated the ability of selected enterocin-producing LAB to suppress *L. monocytogenes* and *Staphylococcus* spp., particularly when applied as adjunct cultures or within multi-hurdle preservation frameworks (Niederhäusern *et al.*, 2020; Nikodinoska *et al.*, 2023).

The effectiveness of enterocin-producing LAB in food systems depends on several key decision criteria. These include strain-level safety verification (absence of virulence and transferable antimicrobial-resistance determinants), compatibility with the target food matrix, stability and activity of enterocins under processing and storage conditions, and minimal impact on sensory attributes. In dairy products, enterocin-producing LAB derived from milk-associated ecosystems have shown particular promise as adjunct cultures during ripening and storage, where they confer a competitive advantage over contaminating pathogens without disrupting desirable fermentation dynamics (Chanos and Williams, 2011; Son *et al.*, 2017).

Application strategy also influences preservation outcomes. Enterocin-producing LAB can be deployed as live protective cultures incorporated into starter formulations or used as sources of purified or semi-purified bacteriocins, depending on regulatory constraints and product characteristics. Their efficacy is often enhanced when combined with complementary hurdles such as refrigeration, vacuum packaging, or

modified-atmosphere storage, reinforcing microbial stability and extending shelf life (Rakhmanova *et al*., 2018; Niederhäusern *et al*., 2020). However, given the dual role of *Enterococcus* spp. in food and clinical contexts, careful strain selection and risk assessment remain essential prerequisites befor application (Sabia *et al*., 2008).

Overall, enterocin-producing LAB represent flexible tools for food biopreservation, offering targeted antimicrobial action while supporting cleaner-label and sustainable preservation strategies. Strategic selection based on safety, functional performance, and technological compatibility is critical to maximize their benefits across different food matrices and processing scenarios. Figure 2 presents a structured flow illustrating the utilization of enterocins in food biopreservation systems, starting from LAB production and purification processes to their application across various food matrices.

Purified, semi-purified and synthetic enterocins

Exploration of enterocin applications has received increasing attention in food safety and preservation, primarily due to their potent antimicrobial activity against Gram-positive pathogens. Enterocins can be categorized into three forms: Mainly purified, semi-purified, and synthetic, each offering unique advantages and applications as food additives or surface-treatment antimicrobial agents (Chikindas *et al*., 2018; Wu *et al*., 2022).

Purified enterocins

Purified enterocins, such as enterocin AS-48 and E-760 have demonstrated strong antimicrobial activity in various food matrices, including dairy and meat products. Enterocin AS-48, for example, has shown a broad antimicrobial spectrum against multiple Gram-positive bacteria, making it a highly promising candidate for food preservation (Popović *et al*., 2024). Similarly, enterocin E-760 has been shown to inhibit key foodborne pathogens; mainly *Salmonella* and *Campylobacter* spp., which are critical concerns in food safety. Enterocin 96 has also proven to be effective in different food model systems, consistently suppressing bacterial growth (Line *et al*., 2008).

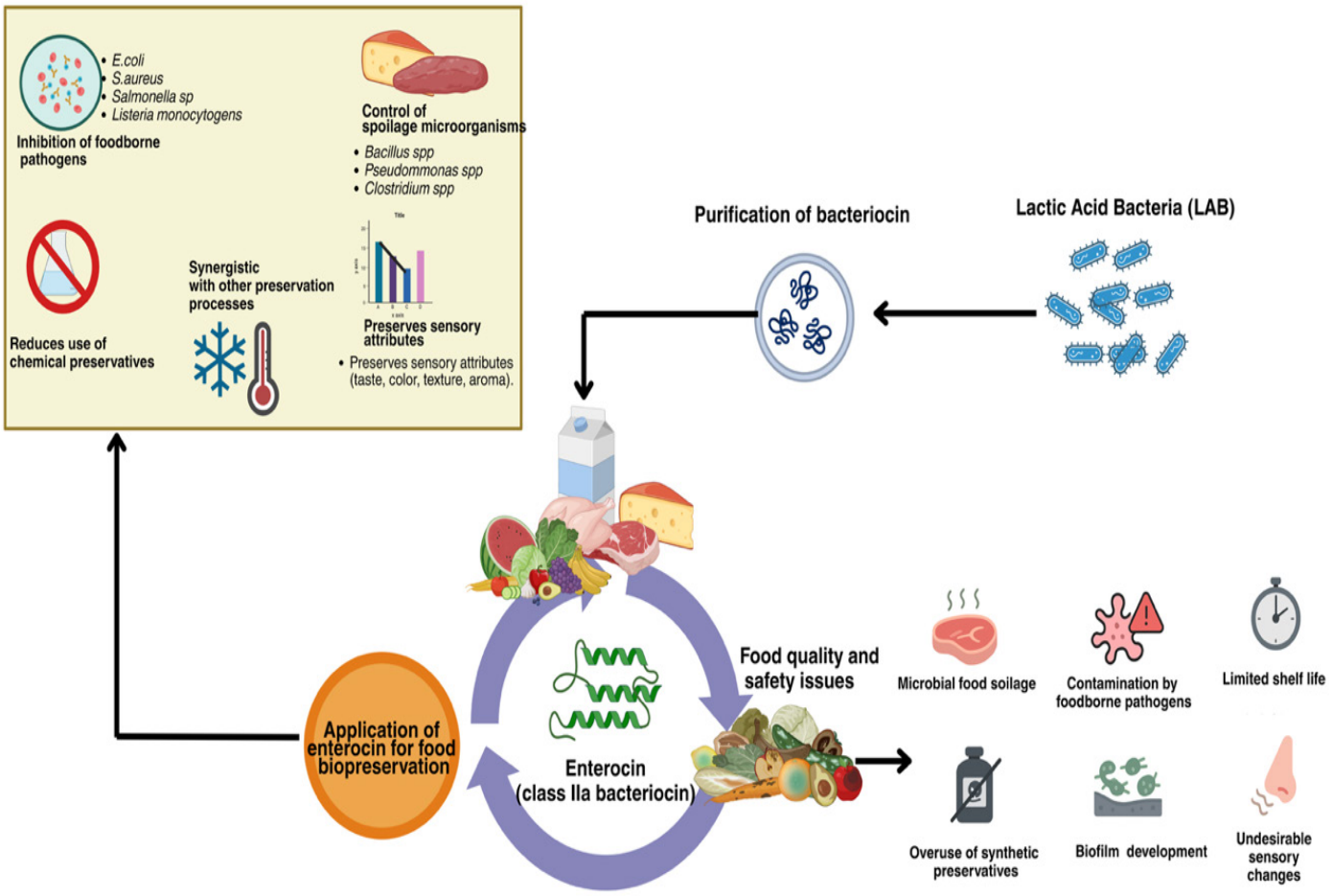

Figure 2: *Schematic workflow illustrating enterocins production and their applications in food biopreservation, adopted by Rameez et al. (2024).*

One substantial advantage of using purified enterocins in food safety is their ability to eliminate concerns associated with introducing live bacterial cultures into food products. This approach enables the incorporation of antimicrobial agents without the potential risk of transferring undesirable or pathogenic strains into the food chain (Popović *et al*., 2024). The inherent stability of purified enterocins further enhances their suitability as food additives, particularly for those applications requiring extended storage and shelf-life preservation.

Semi-purified enterocins

Semi-purified enterocins, such as ESmr18 offer practical advantages for real-world applications, particularly in controlling microbial populations in poultry products. A recent study has shown that applying enterocin ESmr18 to chicken meat considerably reduces *Salmonella* spp. counts during storage, reinforcing its potential as an effective biopreservative (Popović *et al*., 2024). Semi-purification enhances the concentration of active components while preserving the bioactive properties necessary for maintaining antimicrobial activity, without the complexity and high costs associated with full purification. The use of semi-purified enterocins in poultry processing provides dual benefits: improved microbial control and compliance with stringent food safety regulations (Rashid *et al*., 2023). Ultimately, this approach can help increase consumers confidence in the quality and safety of poultry products.

Synthetic enterocins

The development of synthetic enterocins, such as synthetic forms of enterocin A, B, and L50-derived peptides has opened new avenues for research and food applications. Chemical synthesis has enabled the researchers to explore structure–activity relationships and precisely define effective dosages, both of which are essential for designing targeted antimicrobial interventions. Synthetic enterocins maintain remarkable antimicrobial activity while eliminating concerns associated with the pathogenicity or virulence factors that may accompany the use of live bacterial cultures (Salvucci *et al*., 2010).

Notably, certain synthetic variants have demonstrated an enhanced effectiveness against specific pathogens, allowing more targeted approaches to food safety strategies (Ross *et al*., 2020). For instance, the synthetic enterocin CRL35 has shown promising results when used in combination with other antimicrobials, enhancing overall antimicrobial impact and providing a multifaceted strategy for managing foodborne pathogens. Such synergistic combinations highlight the importance of integrating multiple antimicrobial approaches to improve the efficacy against resistant bacteria while minimizing the emergence of new resistance traits in target pathogens (Salvucci *et al*., 2007).

The growing body of research on purified, semi-purified, and synthetic enterocins underscores their substantial potential as natural preservatives in food systems. Purified enterocins exhibit strong inhibitory activity against various Gram-positive pathogens and contribute directly to enhanced food safety, while semi-purified forms offer practical advantages in meat preservation. Meanwhile, synthetic enterocins enable precise dosing and more targeted antimicrobial action. Collectively, these findings highlight the transformative role of enterocins in modern food safety strategies and point out to extensive opportunities for further research and application across the food industry.

Synergy of enerocins with other hurdles and delivery systems

Integration of enterocins into innovative food preservation strategies reflects a shift towards more complex formulations that leverage multiple antimicrobial agents simultaneously. One of the most prominent areas of research in this field is the development of edible films and coatings. Numerous studies have demonstrated that combining essential oils with bacteriocins such as enterocin A enhances antimicrobial effectiveness against several pathogens, including *L. monocytogenes* and *E. coli* (Iseppi *et al*., 2023; Bukvički *et al*., 2023). These combinations allow for localized and gradual release of antimicrobials while reducing the concentrations needed to achieve effective preservation, minimizing the required doses without compromising safety (Iseppi *et al*., 2023).

Edible films and coatings made from biopolymers such as sodium alginate and whey protein have proven effective as barriers against microbial penetration and can be enriched with various natural antimicrobial compounds. Incorporation of essential oils not only contributes to antimicrobial activity but can also improve the mechanical properties of these films (Pavli *et al*., 2019; Rosseto *et al*., 2022). A previous

study conducted by Pavli *et al.* (2019) reported that adding oregano essential oil to sodium alginate films has effectively controlled *L. monocytogenes* on sliced ham when used in combination with high-pressure processing. These findings highlight the synergistic potential of the essential oils, enterocins, and additional preservation hurdles to extend shelf life and enhance food safety.

The growing adoption of combined biopreservation strategies offers several advantages, such as reduced dependence on synthetic preservatives and improved consumers safety profiles. Importantly, the use of biodegradable and ecofriendly packaging materials ensures not only microbial control but also supports sustainability goals (Motelică *et al.*, 2020). Advances in edible films and coatings technologies have enabled the integration of natural antimicrobial compounds, enhancing food safety while aligning with modern ecological demands through reduced plastic wastes (Lucera *et al.*, 2012).

Incorporation of enterocins alongside with other antimicrobials and their integration into advanced delivery systems such as edible films and coatings represents a major step forward in biopreservation strategies. This approach plays a crucial role in improving food safety, extending shelf life, and contributing to healthier and more sustainable food systems. Continued research on specific combinations and their mechanisms of action will be essential to optimize formulations tailored to different food products and storage conditions.

Safety, genomic characterization and regulatory considerations

The growing interests in enterocins derived from *Enterococcus* species reflect their dual potential as probiotic candidates and biopreservative agents. However, safety concerns remain a major barrier to their widespread application, particularly when using viable *Enterococcus* cultures. The key issues include the following:

Virulence factors and opportunistic pathogenicity

Enterococcus faecalis and *E. faecium* are associated with severe healthcare-related infections due to their capacity to harbor a wide range of virulence factors. Studies reported by Jung *et al.* (2017); Hanchi *et al.* (2018) revealed that both clinical and food-derived *Enterococcus* isolates may contain genes encoding aggregation substances, cytolysin, and hyaluronidase components strongly linked to their pathogenicity. The presence of these determinants heightens concerns regarding the application of food-derived enterocin-producing strains, as they may contribute in the dissemination of multidrug-resistance genes and virulence determinants (Hanchi *et al.*, 2018). Safety assessments of enterocin-producing strains isolated from dairy products have indicated that not all strains carry these virulence markers. Several strains have been shown to be free of such genetic determinants, making them more suitable candidates for food applications (Sabia *et al.*, 2008).

Antibiotic resistance and mobile genetic elements

Enterococcus spp. are recognized reservoirs of antibiotic resistance genes, most notably those conferring vancomycin resistance. Previous sesearches reported by Hanchi *et al.* (2018); Terzić-Vidojević *et al.* (2021) identified diverse antibiotic-resistance profiles among enterocin-producing isolates, reinforcing concerns about horizontal gene transfer that could exacerbate antimicrobial resistance in clinical environments. These findings underscore the importance of genomic characterization, as the presence of antimicrobial resistance genes often correlates strongly with virulence profiles, complicating risk assessment for their use in food products.

Need for case-by-case, strain-level evaluation

Rigorous, strain level safety evaluation is essential to fully understand the genetic landscape of enterocin-producing strains. *E. faecium* and *E. mundtii* isolated from colostrum highlight the importance of whole genome sequencing, comprehensive screening for virulence and resistance genes, biogenic amine formation, and detection of *in vitro* toxicity (Terzić-Vidojević *et al.*, 2021). Detailed genomic profiling as demonstrated by Zommiti *et al.* (2022) allows clear differentiation between safe and high-risk strains, ensuring that only strains with acceptable safety parameters are considered for biotechnological applications.

Regulatory constraints on GRAS/QPS species

Current regulatory frameworks such as the Generally Recognized as Safe (GRAS) and Qualified Presumption of Safety (QPS) systems predominantly favor *Lactococcus* spp. and related LAB over *Enterococcus* spp. (Zommiti *et al.*, 2022). This regulatory bias presents a considerable barrier to the acceptance

of *Enterococcus* spp. in food safety applications. Consequently, heterologous production of enterocins in well-characterized GRAS LAB has been proposed as a viable strategy to balance functionality with safety (Terzić-Vidojević *et al*., 2021; Zommiti *et al*., 2022). Given the absence of GRAS status for *Enterococcus* spp., extremely cautious approaches are required, such as the use of purified or synthetic enterocins as food additives combined with strict containment of genetic elements to prevent the co-transfer of virulence or antibiotic-resistance genes.

Future perspectives

The future of enterocin-based biopreservation is shaped by emerging trends in microbiology, synthetic biology, and risk assessment. Understanding these trends is critical for advancing food safety and enhancing the effectiveness of enterocins as biopreservative agents. One of the most promising developments in this area is genome mining and pangenomic analysis, enabling the discovery of novel enterocins gene clusters (Khan *et al*., 2023). These innovations include the identification of unusual cyclic peptides and leaderless peptides with unique antimicrobial spectra and mechanisms of action. Such discoveries not only deepen our understanding of enterocins but also broaden their potential applications in food preservation and safety. By mapping this genetic reservoir, researchers can identify new peptides with enhanced antimicrobial potency, expanding the array of available biopreservative agents.

Advances in synthetic biology and heterologous expression systems in several microbial strains such as *Pichia pastoris* and *Lactococcus lactis* (Hong *et al*., 2025) further enable high-yield production of enterocins. These systems allow researchers to engineer peptides and construct combinatorial libraries designed to optimize stability and specificity. The ability to fine-tune enterocins characteristics through peptide engineering represents a transformative step forward, improving the feasibility and effectiveness of biopreservation strategies. This synthetic approach not only broadens the functional applications of enterocins but also helps addressing challenges related to stability and shelf life when deployed in real food systems.

In parallel with genetic and synthetic advancements, the development of more sophisticated delivery systems provides additional control over the spatial release and targeting of enterocins on food surfaces (Mohanty *et al*., 2025). These innovative delivery mechanisms allow more precise application, ensuring that antimicrobial activity is concentrated where it is most needed without compromising food quality. Such technologies support sustained antimicrobial action, effectively extending product shelf life and enhance protection against microbial contamination.

Risk-benefit modeling and quantitative microbial risk assessment (QMRA) are also emerging as essential tools for regulators and food industry (Magnusson *et al*., 2012). QMRA helps evaluate how enterocins-based hurdles influence the overall risk profile of food products in relation to various pathogens, supporting informed decision-making in food safety management. This scientific approach systematically quantifies risks associated with foodborne pathogens and guides the implementation of enterocins-based biopreservation strategies by clarifying their comparative effectiveness relative to traditional methods.

Collectively, these developments offer new avenues for enhancing food safety and expanding the use of enterocins in preventing microbial contamination of food products. Future research and development will likely build upon these trends to produce more targeted, efficient, and adaptive biopreservation solutions that can meet the evolving demands of modern food industry.

Conclusions and Recommendations

Enterocins are natural antimicrobial peptides that primarily inhibit Gram-positive bacteria by targeting their membranes *via* receptor-mediated mechanisms and inducing rapid cellular dysfunction. Their stability across broad pH ranges, temperatures, and food-processing conditions supports their suitability for biopreservation; however, industrial translation depends on feasibility-driven application choices. In the near term, the most practical approach is the use of purified or semi-purified enterocins as antimicrobial ingredients, followed by their production through controlled fermentation or heterologous expression in well-characterized GRAS/QPS hosts, both of which mitigate biosafety and regulatory concerns associated with viable *Enterococcus* strains. The use of enterocin-producing strains as protective cultures represents a longer-term option and should be restricted to fully genome-characterized isolates lacking transferable

virulence or antimicrobial-resistance determinants. Future developmental studies are recommended to prioritize the standardized safety assessment, optimize enterocins performance in complex food matrices, and their integration into multi-hurdle preservation strategies such as active packaging and cold storage, with validation at pilot and industrial scales to enable reliable commercial adoption.

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Novelty Statement

This review unifies enterocin's producers diversity, gene cluster organization, and membrane-targeting mechanisms with practical deployment options and safety regulatory requirements, providing a translation-focused framework for enterocins-based food biopreservation.

Author's Contribution

RF, LER: Methodology, analysis, and writing. RF, TES, HE: Data analysis, Interpretation, and Reviewing. RF, PS, KUA, AAR: Conceptualization, interpretation, and reviewing. LER: Conceptualization, supervison, and reviewing. All authors contributed to the final manuscript and discussed the outcomes.

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Generative AI and AI-assisted technology statement
During the preparation of this manuscript, the authors used Grammarly solely to improve language clarity and readability. The authors reviewed and verified all content and confirm that all scientific interpretations, data analysis, and conclusions are their own. The authors take full responsibility for the final manuscript.

Conflict of interests
The authors have declared no conflicts of interest.

References

Abanoz, H.S. and Kunduhoglu, B., 2018. Antimicrobial activity of a bacteriocin produced by enterococcus faecalis kt11 against some pathogens and antibiotic-resistant Bacteria. Korean J. Food Sci. Anim. Resour., 38(5): 1064–1079. https://doi.org/10.5851/kosfa.2018.e40

Abengozar, M.A., Cebrian, R., Saugar, J.M., Garate, T., Valdivia, E., Martinez-Bueno, M. and Rivas, L., 2017. Enterocin AS-48 as evidence for the use of bacteriocins as new leishmanicidal agents. Antimicrob. Agents Chemother., 61(4): 10-1128. https://doi.org/10.1128/AAC.02288-16

Agüero, N., Frizzo, L., Ouwehand, A., Aleu, G. and Rosmini, M., 2020. Technological characterisation of probiotic lactic acid bacteria as starter cultures for dry fermented sausages. Foods, 9(5): 596. https://doi.org/10.3390/foods9050596

Alang, H., Kusnadi, J., Ardyati, T. and Suharjono, S., 2020. Optimization and characterization of enterocin enterococcus faecalis k2b1 isolated from toraja's belang buffalo milk, south sulawesi, indonesia. Biodiv. J. Biol. Diver., 21(3). https://doi.org/10.13057/biodiv/d210351

Al-Madboly, L.A., El-Deeb, N.M., Kabbash, A., Nael, M.A., Kenawy, A.M. and Ragab, A.E., 2020. Purification, characterization, identification, and anticancer activity of a circular bacteriocin from *Enterococcus thailandicus*. Front. Bioeng. Biotechnol., 8: 450. https://doi.org/10.3389/fbioe.2020.00450.

Alvarez-Sieiro, P., Montalban-Lopez, M., Mu, D. and Kuipers, O.P., 2016. Bacteriocins of lactic acid bacteria: Extending the family. Appl. Microbiol. Biotechnol., 100(7): 2939–2951. Springer Verlag. https://doi.org/10.1007/s00253-016-7343-9

Anumudu, C.K., Omoregbe, O., Hart, A., Miri, T., Eze, U.A. and Onyeaka, H., 2022. Applications of bacteriocins of lactic acid bacteria in biotechnology and food preservation: A bibliometric review. Open Microbiol. J., 16(1). https://doi.org/10.2174/18742858-v16-e2206300

Ayivi, R.D., Gyawali, R., Krastanov, A., Aljaloud,

S.O., Worku, M., Tahergorabi, R., da Silva, RC., Ibrahim, S.A., 2020. Lactic acid bacteria: Food safety and human health applications. Dairy, 1(3): 202-232. https://doi.org/10.3390/dairy1030015

Bansal, P., Kumar, R., Dhanda, S., 2022. Characterization of starter cultures and nutritional properties of pediococcus acidilactici ncdc 252: a potential probiotic of dairy origin. J. Food Process. Preserv., 46(10). https://doi.org/10.1111/jfpp.16817

Belgacem, Z., Abriouel, H., Omar, N., López, R., Martı-nez-Cañamero, M., Gálvez, A. and Manaï, M., 2010. Antimicrobial activity, safety aspects, and some technological properties of bacteriocinogenic enterococcus faecium from artisanal tunisian fermented meat. Food Contr., 21(4): 462-470. https://doi.org/10.1016/j.foodcont.2009.07.007

Belguesmia, Y., Choiset, Y., Prévost, H., Dalgalarrondo, M., Chobert, J. and Drider, D., 2010. Partial purification and characterization of the mode of action of enterocin s37: A bacteriocin produced byenterococcus faecaliss37 isolated from poultry feces. J. Environ. Publ. Health, 2010: 1-8. https://doi.org/10.1155/2010/986460

Bhattacharya, D., Nanda, P.K., Pateiro, M., Lorenzo, J.M., Dhar, P., Das, A.K., 2022. Lactic acid bacteria and bacteriocins: Novel biotechnological approach for biopreservation of meat and meat products. Microorganisms, 10(10):2058. https://doi.org/10.3390/microorganisms10102058

Birri, D., Brede, D., Forberg, T., Holo, H. and Nes, I., 2010. Molecular and genetic characterization of a novel bacteriocin locus in enterococcus avium isolates from infants. Appl. Environ. Microbiol., 76(2): 483-492. https://doi.org/10.1128/AEM.01597-09

Bukvički, D., D'Alessandro, M., Rossi, S., Siroli, L., Gottardi, D., Braschi, G., Patrignani, F. and Lanciotti, R., 2023. Essential oils and their combination with lactic acid bacteria and bacteriocins to improve the safety and shelf life of foods: A review. Foods, 12(17): 3288. https://doi.org/10.3390/foods12173288

Chanos, P. and Williams, D., 2011. Anti-listeria bacteriocin-producing bacteria from raw ewe's milk in northern greece. J. Appl. Microbiol., 110(3): 757-768. https://doi.org/10.1111/j.1365-2672.2010.04932.x

Chikindas, M.L., Weeks, R., Drider, D., Chistyakov, V.A. and Dicks, L.M., 2018. Functions and emerging applications of bacteriocins. Curr. Opin. Biotechnol., 49: 23–28. Elsevier Ltd. https://doi.org/10.1016/j.copbio.2017.07.011

Cintas, L.M., Casaus, P., Herranz, C., Håvarstein, L.S., Holo, H., Hernández, P.E., Holo, H., Hernadedez, P.E. and Nes, I., 2000. Biochemical and genetic evidence that enterococcus faecium l50 produces enterocins l50a and l50b, the sec-dependent enterocin p, and a novel bacteriocin secreted without an n-terminal extension termed enterocin q. J. Bacteriol., 182(23): 6806-6814. https://doi.org/10.1128/JB.182.23.6806-6814.2000

Cirat, R., Capozzi, V., Benmechernene, Z., Spano, G., Grieco, F. and Fragasso, M., 2024. Lab antagonistic activities and their significance in food biotechnology: Molecular mechanisms, food targets, and other related traits of interest. Fermentation, 10(4): 222. https://doi.org/10.3390/fermentation10040222

Costa, R., Voloski, F., Mondadori, R., Duval, E. and Fiorentini, Â., 2019. Preservation of meat products with bacteriocins produced by lactic acid bacteria isolated from meat. J. Food Qual., 2019: 1-12. https://doi.org/10.1155/2019/4726510

Darbandi, A., Asadi, A., Mahdizade Ari, M., Ohadi, E., Talebi, M., Halaj Zadeh, M., Darb Emamie, A., Ghanavati, R. and Kakanj, M., 2022. Bacteriocins: Properties and potential use as antimicrobials. J. Clin. Lab. Anal., 36(1): 1–40. John Wiley and Sons Inc. https://doi.org/10.1002/jcla.24093

Ennahar, S., Sashihara, T., Sonomoto, K. and Ishizaki, A., 2000. Class iia bacteriocins: biosynthesis, structure and activity. Fems Microbiol. Rev., 24(1): 85-106. https://doi.org/10.1111/j.1574-6976.2000.tb00534.x

Franz, C.M., Van Belkum, M.J., Holzapfel, W.H., Abriouel, H. and Gálvez, A., 2007. Diversity of enterococcal bacteriocins and their grouping in a new classification scheme. FEMS Microbiol. Rev., 31(3): 293-310. https://doi.org/10.1111/j.1574-6976.2007.00064.x

Garcia-Vela, S., Guay, L.D., Rahman, M.R.T., Biron, E., Torres, C. and Fliss, I., 2024. Antimicrobial activity of synthetic enterocins A, B, P, SEK4, and L50, alone and in combinations, against Clostridium perfringens. Int. J. Mol. Sci., 25(3):

1597. https://doi.org/10.3390/ijms25031597

Garmasheva, I. and Oleschenko, L., 2023. Screening of bacteriocin-producing dairy enterococcus strains using low-cost culture media. Front. Microbiol., 14. https://doi.org/10.3389/fmicb.2023.1168835

Gupta, A., Tiwari, S.K., Netrebov, V. and Chikindas, M.L., 2016. Biochemical properties and mechanism of action of enterocin LD3 purified from Enterococcus hirae LD3. Probiot. Antimicrob. Proteins, 8(3): 161-169. https://doi.org/10.1007/s12602-016-9217-y

Hanchi, H., Hammami, R., Fernandez, B., Kourda, R., Hamida, J. and Fliss, I., 2016. Simultaneous production of formylated and nonformylated enterocins l50a and l50b as well as 61a, a new glycosylated durancin, by enterococcus durans 61a, a strain isolated from artisanal fermented milk in tunisia. J. Agric. Food Chem., 64(18): 3584-3590. https://doi.org/10.1021/acs.jafc.6b00700

Hanchi, H., Mottawea, W., Sebei, K. and Hammami, R., 2018. The genus enterococcus: Between probiotic potential and safety concerns an update. Front. Microbiol., 9. https://doi.org/10.3389/fmicb.2018.01791

Hernández-González, J.C., Martínez-Tapia, A., Lazcano-Hernández, G., García-Pérez, B.E. and Castrejón-Jiménez, N.S., 2021. Bacteriocins from lactic acid bacteria. A powerful alternative as antimicrobials, probiotics, and immunomodulators in veterinary medicine. Animals, 11(4): 979. https://doi.org/10.3390/ani11040979

Herranz, C. and Driessen, A.J., 2005. Sec-mediated secretion of bacteriocin enterocin P by *Lactococcus lactis*. Appl. Environ. Microbiol., 71(4): 1959-1963. https://doi.org/10.1128/AEM.71.4.1959-1963.2005

Hill, D., O'Connor, P., Altermann, E., Day, L., Hill, C., Stanton, C. and Ross, R., 2020. Extensive bacteriocin gene shuffling in the streptococcus bovis/streptococcus equinus complex reveals gallocin d with activity against vancomycin resistant enterococci. Sci. Rep., 10(1). https://doi.org/10.1038/s41598-020-70328-z

Hong, K., Rong, Y., Jiang, Y., Yuan, J., Wu, F., Gao, L. and Wu, X., 2025. Different strategies for the biosynthesis of bioactive peptide using bioengineering technology in Pichia pastoris: A review. Food Bioproc. Technol., 18(5): 4326-4344. https://doi.org/10.1007/s11947-025-03755-3

Iseppi, R., Camellini, S., Zurlini, C., Cigognini, I., Cannavacciuolo, M. and Messi, P., 2023. Essential oils and bacteriocin-based active edible coating: An innovative, natural and sustainable approach for the control of listeria monocytogenes in seafoods. Appl. Sci., 13(4): 2562. https://doi.org/10.3390/app13042562

Izquierdo, E., Wagner, C., Marchioni, E., Aoude-Werner, D. and Ennahar, S., 2009. Enterocin 96, a novel class II bacteriocin produced by Enterococcus faecalis WHE 96, isolated from Munster cheese. Appl. Environ. Microbiol., 75(13): 4273-4276. https://doi.org/10.1128/AEM.02772-08

Jaouani, I., Abbassi, M.S., Alessandria, V., Bouraoui, J., Ben Salem, R., Kilani, H. and Cocolin, L., 2014. High inhibition of Paenibacillus larvae and Listeria monocytogenes by Enterococcus isolated from different sources in Tunisia and identification of their bacteriocin genes. Lett. Appl. Microbiol., 59(1): 17-25. https://doi.org/10.1111/lam.12239

Javed, A., Masud, T., Ain, Q., Imran, M. and Maqsood, S., 2011. Enterocins of enterococcus faecium, emerging natural food preservatives. Ann. Microbiol., 61(4): 699-708. https://doi.org/10.1007/s13213-011-0223-8

Javed, I., Ahmed, S., Ali, M., Ahmad, B., Ghumro, P., Hameed, A. and Chaudry, G., 2010. Bacteriocinogenic potential of newly isolated strains of enterococcus faecium and enterococcus faecalis from dairy products of pakistan. J. Microbiol. Biotechnol., 20(1): 153-160. https://doi.org/10.4014/jmb.0904.04024

Jin, G., Zhang, M., Wang, X., Zhang, Y., Jiang, G. and Mei, L., 2025. Characteristics of exopolysaccharides egg white protein composite gel and its application in low fat sausage. Food Chem.X, 26: 102290. https://doi.org/10.1016/j.fochx.2025.102290

Jung, A., Metzner, M. and Ryll, M., 2017. Comparison of pathogenic and non-pathogenic enterococcus cecorum strains from different animal species. BMC Microbiol., 17(1). https://doi.org/10.1186/s12866-017-0949-y

Kasimin, M.E., Shamsuddin, S., Molujin, A.M., Sabullah, M.K., Gansau, J.A. and Jawan, R., 2022. Enterocin: Promising biopreservative produced by *Enterococcus*

sp. Microorganisms, 10(4): 684. https://doi.org/10.3390/microorganisms10040684

Khan, K., Jalal, K. and Uddin, R., 2023. Pangenome profiling of novel drug target against vancomycin-resistant *Enterococcus faecium*. J. Biomol. Struct. Dyn., 41(24): 15647-15660. https://doi.org/10.1080/07391102.2023.2191134

Kurushima, J., Nakane, D., Nishizaka, T. and Tomita, H., 2014. Bacteriocin protein bacl1of enterococcus faecalis targets cell division loci and specifically recognizes l-ala2-cross-bridged peptidoglycan. J. Bacteriol., 197(2): 286-295. https://doi.org/10.1128/JB.02203-14

Ladjouzi, R., Lucau-Danila, A., Benachour, A. and Drider, D., 2020. A leaderless two-peptide bacteriocin, enterocin dd14, is involved in its own self-immunity: Evidence and insights. Front. Bioeng. Biotechnol., 8. https://doi.org/10.3389/fbioe.2020.00644

Line, J., Ea, S., Eruslanov, B., Perelygin, V., Mitsevich, E., Mitsevich, I., Levchuk, V.P., Svetoch, O.E., Seal, B. S., Siragusa, G.R. and Stern, N., 2008. Isolation and purification of enterocin e-760 with broad antimicrobial activity against gram-positive and gram-negative bacteria. Antimicrob. Agents Chemother., 52(3): 1094-1100. https://doi.org/10.1128/AAC.01569-06

Liu, G., Griffiths, M., Wu, P., Wang, H., Zhang, X. and Li, P., 2011. Enterococcus faecium lm-2, a multi-bacteriocinogenic strain naturally occurring in byaslag, a traditional cheese of inner mongolia in china. Food Contr., 22(2): 283-289. https://doi.org/10.1016/j.foodcont.2010.07.023

Liu, G., Wang, Y., Li, X., Xu, H., Xu, D., Zhou, Y., Mehmood, A. and Wang, C., 2019. Genetic and biochemical evidence that enterococcus faecalis gr17 produces a novel and sec-dependent bacteriocin, enterocin gr17. Front. Microbiol., 10. https://doi.org/10.3389/fmicb.2019.01806

Liu, X., Vederas, J., Whittal, R., Zheng, J., Stiles, M., Carlson, D., Franz, C.M.A.P. and Belkum, M., 2011. Identification of an n-terminal formylated, two-peptide bacteriocin from enterococcus faecalis 710c. J. Agric. Food Chem., 59(10): 5602-5608. https://doi.org/10.1021/jf104751v

Lopes, J., Lencastre, H. and Conceição, T., 2024. Genomic analysis of enterococcus faecium from non-clinical settings: Antimicrobial resistance, virulence, and clonal population in livestock and the urban environment. Front. Microbiol., 15. https://doi.org/10.3389/fmicb.2024.1466990

Lucera, A., Costa, C., Conte, A. and Nobile, M., 2012. Food applications of natural antimicrobial compounds. Front. Microbiol., 3. https://doi.org/10.3389/fmicb.2012.00287

Luenglusontigit, P., Sathapondecha, P., Saengsuwan, P., Surachat, K., Boonserm, P. and Singkhamanan, K., 2023. Effects of postbiotic from bacteriocin-like inhibitory substance producing *Enterococcus faecalis* on toxigenic Clostridioides difficile. J. Health Sci. Med. Res., 41(4): 2023918. https://doi.org/10.31584/jhsmr.2023918

Magnússon, S.H., Gunnlaugsdóttir, H., Van Loveren, H., Holm, F., Kalogeras, N., Leino, O., Luteijn, J.M., Odekerken, G., Pohjola, M.V., Tijhuis, M.J., Tuomisto, J.T., Ueland, O., White, B.C. and Verhagen, H., 2012. State of the art in benefit–risk analysis: Food microbiology. Food Chem. Toxicol., 50(1): 33-39. https://doi.org/10.1016/j.fct.2011.06.005

Mareková, M., Lauková, A., Vuyst, L., Skaugen, M. and Nes, I., 2003. Partial characterization of bacteriocins produced by environmental strain enterococcus faecium ek13. J. Appl. Microbiol., 94(3): 523-530. https://doi.org/10.1046/j.1365-2672.2003.01861.x

Martínez-Bueno, M., Maqueda, M., Gálvez, A., Samyn, B., Beeumen, J., Coyette, J. and Valdivia, E., 1994. Determination of the gene sequence and the molecular structure of the enterococcal peptide antibiotic as-48. J. Bacteriol., 176(20): 6334-6339. https://doi.org/10.1128/jb.176.20.6334-6339.1994

Merzoug, M., Bendida, K., Aireche, M., Zater, Z.Y., Brakna, C.N., Hammadi, A.I. and Saidi, D., 2025. Isolation and characterization of enterocin-producing enterococcus faecium strains from algerian traditional food "dried figs marinated in olive oil": Functional and safety evaluations. Foods, 14(5): 766. https://doi.org/10.3390/foods14050766

Mohanty, D., Suar, M. and Panda, S.K., 2025. Nanotechnological interventions in bacteriocin formulations–advances, and scope for challenging food spoilage bacteria and drug-resistant foodborne pathogens. Crit. Rev. Food Sci. Nutr., 65(6): 1126-1143. https://doi.org/10.1080/10408398.2023.2289184

Motelică, L., Ficai, D., Ficai, A., Oprea, O., Kaya, D. and Andronescu, E., 2020. Biodegradable antimicrobial food packaging: Trends and perspectives. Foods, 9(10): 1438. https://doi.org/10.3390/foods9101438

Nachér-Vázquez, M., Ruiz-Masó, J., Mohedano, M., Solar, G., Aznar, R. and López, P., 2017. Dextransucrase expression is concomitant with that of replication and maintenance functions of the pmn1 plasmid in lactobacillus sakei mn1. Front. Microbiol., 8. https://doi.org/10.3389/fmicb.2017.02281

Ness, I.F., Diep, D.B. and Ike, Y., 2014. Enterococcal bacteriocins and antimicrobial proteins that contribute to niche control. In: Gilmore MS, Clewell DB, Ike Y, *et al*., editors. Enterococci: From Commensals to Leading Causes of Drug Resistant Infection [Internet]. Boston: Massachusetts Eye and Ear Infirmary.

Niederhäusern, S., Camellini, S., Sabia, C., Iseppi, R., Bondi, M. and Messi, P., 2020. Antilisterial activity of bacteriocins produced by lactic bacteria isolated from dairy products. Foods, 9(12): 1757. https://doi.org/10.3390/foods9121757

Nikodinoska, I., Tabanelli, G., Baffoni, L., Gardini, F., Gaggìa, F., Barbieri, F. and Gioia, D., 2023. Characterization of lactic acid bacteria isolated from spontaneously fermented sausages: Bioprotective, technological and functional properties. Foods, 12(4): 727. https://doi.org/10.3390/foods12040727

Oladipo, I., Sanni, A., Chakraborty, W., Chakravorty, W., Jana, S., Rudra, D. and Swarnakar, S., 2015. Technological properties of strains of enterococcus gallinarum isolated from selected nigerian traditional fermented foods. Malays. J. Microbiol., 11: 1-13. https://doi.org/10.21161/mjm.64014

Park, S., Itoh, K. and Fujisawa, T., 2003. Characteristics and identification of enterocins produced by enterococcus faecium jcm 5804t. J. Appl. Microbiol., 95(2): 294-300. https://doi.org/10.1046/j.1365-2672.2003.01975.x

Pavli, F., Argyri, A., Skandamis, P., Nychas, G., Tassou, C. and Chorianopoulos, N., 2019. Antimicrobial activity of oregano essential oil incorporated in sodium alginate edible films: Control of listeria monocytogenes and spoilage in ham slices treated with high pressure processing. Materials, 12(22): 3726. https://doi.org/10.3390/ma12223726

Popović, N., Veljović, K., Radojević, D., Brdarić, E., Stevanović, D., Živković, M. and Kojić, M., 2024. Insight into the probiogenomic potential of enterococcus faecium bgpas 1-3 and application of a potent thermostable bacteriocin. Foods, 13(16): 2637. https://doi.org/10.3390/foods13162637

Prajapati, N., Patel, J., Singh, S., Yadav, V., Joshi, C., Patani, A., Prajapati, D., Sahoo, D.K. and Patel, A., 2023. Postbiotic production: Harnessing the power of microbial metabolites for health applications. Front. Microbiol., 14. https://doi.org/10.3389/fmicb.2023.1306192

Putri, D.A., Lei, J., Rossiana, N. and Syaputri, Y., 2024. Biopreservation of food using bacteriocins from lactic acid bacteria: Classification, mechanisms, and commercial applications. Int. J. Microbiol., 2024(1): 8723968. https://doi.org/10.1155/ijm/8723968

Rakhmanova, A., Khan, Z. and Shah, K., 2018. A mini review fermentation and preservation: Role of lactic acid bacteria. Moj Food Process. Technol., 6(5): 414-417. https://doi.org/10.15406/mojfpt.2018.06.00197

Rameez, K.M., Santhoshkumar, P., Yoha, K.S. and Moses, J.A., 2024. Biopreservation of food using probiotics: Approaches and challenges. Curr. Res. Nutr. Food Sci. J., 12(2): 539-560. https://doi.org/10.12944/CRNFSJ.12.2.5

Rashid, M., Sharma, S., Kaur, A., Kaur, A. and Kaur, S., 2023. Biopreservative efficacy of Enterococcus faecium-immobilised film and its enterocin against Salmonella enterica. AMB Express, 13(1): 11. https://doi.org/10.1186/s13568-023-01516-z

Ross, J., Fields, F., Kalwajtys, V., Gonzalez, A., O'Connor, S., Zhang, A., Moran, T.E., Hammers, D.E., Carothers, K.E. and Lee, S., 2020. Synthetic peptide libraries designed from a minimal alpha-helical domain of as-48-bacteriocin homologs exhibit potent antibacterial activity. Front. Microbiol., 11. https://doi.org/10.3389/fmicb.2020.589666

Rosseto, M., Rigueto, C., Alessandretti, I., Oliveira, R., Wohlmuth, D., Loss, R., Dettmer, A. and Richards, N., 2022. Whey-based polymeric films for food packaging applications: A review of recent trends. J. Sci. Food Agric., 103(7): 3217-3229. https://doi.org/10.1002/jsfa.12310

Sabia, C., Niederhäusern, S., Guerrieri, E., Messi,

P., Anacarso, I., Manicardi, G. and Bondi, M., 2008. Detection of bacteriocin production and virulence traits in vancomycin-resistant enterococci of different sources. J. Appl. Microbiol., 104(4), 970-979. https://doi.org/10.1111/j.1365-2672.2007.03612.x

Salvucci, E., Hébert, E., Sesma, F. and Saavedra, L., 2010. Combined effect of synthetic enterocin crl35 with cell wall, membrane-acting antibiotics and muranolytic enzymes against listeria cells. Lett. Appl. Microbiol., 51: 191-195. https://doi.org/10.1111/j.1472-765X.2010.02879.x

Salvucci, E., Saavedra, L. and Sesma, F., 2007. Short peptides derived from the nh2-terminus of subclass iia bacteriocin enterocin crl35 show antimicrobial activity. J. Antimicrob. Chemother., 59(6): 1102-1108. https://doi.org/10.1093/jac/dkm096

Sanchez, J., Borrero, J., Gómez- Sala, B., Basanta, A., Herranz, C., Cintas, L. and Hernández, P., 2008. Cloning and heterologous production of hiracin jm79, a sec-dependent bacteriocin produced by enterococcus hirae dch5, in lactic acid bacteria and pichia pastoris. Appl. Environ. Microbiol., 74(8): 2471-2479. https://doi.org/10.1128/AEM.02559-07

Settanni, L., Guarcello, R., Gaglio, R., Francesca, N., Aleo, A., Felis, G.E. and Moschetti, G., 2014. Production, stability, gene sequencing and in situ anti-Listeria activity of mundticin KS expressed by three *Enterococcus mundtii* strains. Food Contr., 35(1): 311-322. https://doi.org/10.1016/j.foodcont.2013.07.022

Son, S., Jeon, H., Yang, S., Sim, M., Kim, Y., Lee, N. and Paik, H., 2017. Probiotic lactic acid bacteria isolated from traditional korean fermented foods based on β-glucosidase activity. Food Sci. Biotechnol., 27(1): 123-129. https://doi.org/10.1007/s10068-017-0212-1

Souza, N., Carvalho, L., Nogueira, M., Furlaneto, M. and Furlaneto-Maia, L., 2024. Potential of enterocin from enterococcus durans mf5 in controlling listeria species. J. Dairy Res., 91(4): 516-524. https://doi.org/10.1017/S0022029925000160

Terzić-Vidojević, A., Veljović, K., Popović, N., Tolinački, M. and Golić, N., 2021. Enterococci from raw-milk cheeses: Current knowledge on safety, technological, and probiotic concerns. Foods, 10(11): 2753. https://doi.org/10.3390/foods10112753

Toğay, S., Ay, M., Güneşer, O. and Yüceer, Y., 2016. Investigation of antimicrobial activity and enta and entb genes in enterococcus faecium and enterococcus faecalis strains isolated from naturally fermented turkish white cheeses. Food Sci. Biotechnol., 25(6): 1633-1637. https://doi.org/10.1007/s10068-016-0251-z

Tuncer, B., Ay, Z. and Tuncer, Y., 2013. Occurrence of enterocin genes, virulence factors, and antibiotic resistance in 3 bacteriocin-producer enterococcus faecium strains isolated from turkish tulum cheese. Turk. J. Biol., 37: 443-449. https://doi.org/10.3906/biy-1209-26

Turhan, E., Erginkaya, Z., Türemiş, A. and Terkuran, M., 2020. Antibacterial effect of bacteriocinogenic enterococci from different sources on listeria monocytogenes. Tarım Bilimleri Dergisi.

Vimont, A., Fernandez, B., Hammami, R., Ababsa, A., Daba, H. and Fliss, I., 2017. Bacteriocin-producing enterococcus faecium lcw 44: A high potential probiotic candidate from raw camel milk. Front. Microbiol., 8. https://doi.org/10.3389/fmicb.2017.00865

Wang, R.X., Li, Y.Q., Sun, G.J., Wang, C.Y., Liang, Y., Hua, D.L., Chen, L. and Mo, H.Z., 2023. The improvement and mechanism of gelation properties of mung bean protein treated by ultrasound. LWT, 182: 1–10. https://doi.org/10.1016/j.lwt.2023.114811

Wang, Y., Shang, N., Huang, Y., Gao, B. and Li, P., 2024. The progress of the biotechnological production of class IIa bacteriocins in various cell factories and its future challenges. Int. J. Mol. Sci., 25(11): 5791. https://doi.org/10.3390/ijms25115791

Wu, Y., Pang, X., Wu, Y., Liu, X. and Zhang, X., 2022. Enterocins: Classification, synthesis, antibacterial mechanisms and food applications. Molecules, 27(7): 2258. https://doi.org/10.3390/molecules27072258

Yap, P.G., Lai, Z.W. and Tan, J.S., 2022. Bacteriocins from lactic acid bacteria: Purification strategies and applications in food and medical industries: A review. Beni-Suef Univ. J. Basic Appl. Sci., 11(1): 51. https://doi.org/10.1186/s43088-022-00227-x

Yordanova, R., Hristova, P. and Platikanova, M., 2024. Benefits and risks of using *Enterococcus* species in food or as probiotics. J. IMAB, 30(2): 5569-5573. https://doi.org/10.5272/

jimab.2024302.5569

Zhadyra, S., Tao, F. and Xu, P., 2025. Two-dimensional gc–tofms analysis of volatile organic compounds in fermented camel milk (shubat). Foods, 14(17): 2995. https://doi.org/10.3390/foods14172995

Zommiti, M., Cambronel, M., Maillot, O., Barreau, M., Sebei, K., Feuilloley, M., Ferchichi, M. and Connil, N., 2018. Evaluation of probiotic properties and safety of enterococcus faecium isolated from artisanal tunisian meat "dried ossban". Front. Microbiol., 9. https://doi.org/10.3389/fmicb.2018.01685

Zommiti, M., Chevalier, S., Feuilloley, M. and Connil, N., 2022. Special issue enterococci for probiotic use: Safety and risk: Editorial. Microorganisms, 10(3): 604. https://doi.org/10.3390/microorganisms10030604