Characteristics of Heterologous Plantaricin from *Lactobacillus plantarum* and its Future in Food Preservation

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**ABSTRACT**

*Lactobacillus plantarum*, a lactic acid bacterium, produces organic acids, fatty acids, ammonia, hydrogen peroxide, diacetyl, and bacteriocin to survive in an unfavorable environment. Plantaricin, a class II bacteriocin produced by *L. plantarum*, is reported to be heterologous, and its ability to inhibit or kill pathogenic bacteria is very broad, with potential application as a bio-preservative. Plantaricin production is regulated by genetically organized operons, which also encode structural genes, immunity proteins, and secretion genes in plasmids or chromosomes. The mechanism of action against pathogenic bacteria depends on the characteristics of plantaricin. The most common bactericidal mechanisms are disruption of the cell wall integrity and inhibition of protein or nucleic acid synthesis. This review focuses on characterization of the heterologous mechanisms of plantaricin to inhibit and kill pathogenic bacteria and the future role of plantaricin for food preservation. With this review, we hope to contribute to innovation in food preservation, by promoting a better understanding of this natural resource.

**Keywords**

class II bacteriocin, *Lactobacillus plantarum*, plantaricin

1. **Introduction**

*Lactobacillus plantarum* is a lactic acid bacterium found in nutrient-rich environments such as plants, meat, fish, and dairy products. *L. plantarum* produces organic acids, fatty acids, ammonia, hydrogen peroxide, diacetyl, and bacteriocin as well as other substances to grow and survive in its environment (Azizi *et al.*, 2017). The bacteriocin produced by *L. plantarum* is known as plantaricin and is generally reported as a class II bacteriocin, a very broad class with a variety of bactericidal/bacteriostatic mechanisms (Ahmad *et al.*, 2017). Class II bacteriocins are small peptide (< 10 kDa), heat-stable molecules with isoelectric points varying from 8.3 to 10.0 (Todorov, 2009). They have an amphiphilic helical structure that allows for their insertion into the cytoplasmic membrane of the target cell, thereby promoting membrane depolarization and cell death (Diep *et al.*, 2007).

The uses of plantaricin are widely studied and has developed rapidly, with diverse applications such as antibacterial packaging film (Yang *et al.*, 2019), reduction of intestinal cancer cells (De Giani *et al.*, 2019), bio-preservation of fresh fish and shellfish (Čanak *et al.*, 2018), extending the shelf-life of food without altering the nutritional quality of products (Flórez and Mayo, 2018), anti-cancer drugs (Baindara *et al.*, 2018), and active polyvinylidene chloride (PVDC) films as antimicrobial wrapping for fresh pork (Xie *et al.*, 2018). Plantaricin...
products can lower cholesterol, act as antioxidants (Devi and Halami, 2019), and are active against Candida (Sharma and Srivastava, 2014). However, most reports concentrate on plantaricin as a bio-preservative, owing to its use in the extension of shelf life and effectiveness against a range of harmful bacteria.

Plantaricin from L. plantarum has been widely reported, and the broad, heterologous nature of plantaricin has been emphasized by several authors. Differences in structural amino acids result in different characteristics of plantaricins, such as resistance to pH and temperature (Tenea and Pozo, 2019), and antimicrobial activity (Wang et al., 2018). Differences are also influenced by the location of the plantaricin-coding genes (Todorov, 2009). In L. plantarum, these are located in operon clusters, which may be located on chromosomes, plasmids, or transposons (Malik et al., 2016; Todorov, 2009). The mechanism used to inhibit and kill pathogenic bacteria depends on the characteristics of the plantaricin; different classes have different mechanisms. Commonly, plantaricin disrupts cell wall integrity and inhibits protein or nucleic acid synthesis (Ahmad et al., 2017). It has been reported that the bacterial membrane is the target of bacteriocins (Diep et al., 2001); therefore, it is worth summarizing and clarifying the mechanisms underlying the bactericidal/bacteriostatic activity of plantaricin against pathogenic bacteria.

In this review, we present information about the diversity, characterization, and heterologous expression of plantaricin from L. plantarum that has many benefits to human life, with the aim of promoting its use and stimulating innovation in a diverse range of industries, especially the food industry for food preservation.

2. Classification of bacteriocin Class II

The bacteriocin produced by L. plantarum is known as plantaricin and is usually reported as a class II bacteriocin. These classes have been defined according to biochemical and genetic characterization. Class II bacteriocins are small peptide (<10 kDa), heat-stable, hydrophobic molecules, with amphiphilic α-helical structures (Heeney et al., 2019) and isoelectric points in the range of 8.3–10.0 (Todorov, 2009). Plantaricin has been reported to have high specificity for some clinically pathogenic bacteria and is active against multi-antibiotic resistant strains (Perez et al., 2014). A summary of class II bacteriocins is provided in Table 1.

Different subclasses of bacteriocin have different characteristics, and these characteristics have different mechanisms to inhibit and kill bacteria and provide benefits to the food industry. Plantaricin is a class II bacteriocin that can provide antimicrobial activity at relatively high temperatures (Hata et al., 2010), in highly acidic or alkaline conditions (Rumjuankiat et al., 2015) and at high levels of salinity (Hurtado et al., 2011). Thus, processing conditions need to be considered when applying the knowledge of bacteriocins to industrial applications.

3. Characterization and heterogeneity of plantaricin

Bacteriocin production is regulated by genetically organized operons, usually comprising structural genes and genes encoding proteins responsible for post-translational modification and export (Malik et al., 2016; Van Reenen et al., 2003). Differences in location may provide different structural genes, immunity proteins, and secretion genes. Bacteriocin producers are constantly protected from their own bacteriocins by so-called immunity proteins, whose genetic determinants are usually found within the bacteriocin locus (Malik et al., 2016; Miller et al., 2005). Some plantaricins encode bacteriocin immunity proteins and secretion genes directly, e.g., on L. plantarum WCFS1 is shown in Fig. 1. Different strains produce different plantaricin structural genes. Genes encoding plantaricin are located both on a plasmid and in a chromosomal region. This information explains the diversity and characterization of plantaricin in Table 2.
References

Chikindas et al., 1993; Kaur and Kaur, 2015; Zacharof and Lovitt, 2012

Bengtsson et al., 2020; Kristiansen et al., 2005; Oppegård et al., 2016; Xu et al., 2019

Ahmad et al., 2017; Collins et al., 2017; Diep et al., 2007

Hata et al., 2010; Perez et al., 2014; Todorov, 2009

Application

Natural food preservation
Antibiotics candidate, human clinical drug
Antibiotics candidate

Advantages

Active inhibit various bacteria causes food-borne listeriosis by binding the cytoplasmic membrane of target cells and forming voltage-independent pores which cause an efflux of important cellular metabolites. High efficiency, no drug resistance, have 103 times more active when combined with their complementary peptide than individually and easily amenable through bioengineering to increase either their activity or specificity towards target microorganisms. It can be used as bacteriocin–receptor complex and employ a similar mechanism to target susceptible cells. Heat stable and wide range of pH condition, especially in neutral and alkaline pH

Characteristics

Class IIa is well-known as pediocin-like bacteriocins, antimicrobial spectrum of this subclass is different and strongly active against Listeria spp., share a conserved amino acid sequence -YGNGV- in their structure, but the function of this sequence is still unclear. 

Class IIb is well-known as two peptides bacteriocin, some bacteriocins need both peptides to active, and generally act synergistically. Hydrophobic and amphiphilic α-helices profile. Stable at high temperature and extreme pH.

Small and heat stable peptide, divided into two groups; thiolbiotics and cystibiotics, and N- and C-termini are covalently linked.

Class IIId is still unclassified bacteriocins, all of the bacteriocins which do not include the criteria of the previous three Class II, including sec-dependent bacteriocins and leaderless bacteriocins.

Table 1 Classification of Bacteriocin

| Class II | Characteristics | Advantages | Application | References |
|----------|-----------------|------------|-------------|------------|
| Class IIa | Class IIa is well-known as pediocin-like bacteriocins, antimicrobial spectrum of this subclass is different and strongly active against Listeria spp., share a conserved amino acid sequence -YGNGV- in their structure, but the function of this sequence is still unclear. | Active inhibit various bacteria causes food-borne listeriosis by binding the cytoplasmic membrane of target cells and forming voltage-independent pores which cause an efflux of important cellular metabolites | Natural food preservation | Chikindas et al., 1993; Kaur and Kaur, 2015; Zacharof and Lovitt, 2012 |
| Class IIb | Class IIb is well-known as two peptides bacteriocin, some bacteriocins need both peptides to active, and generally act synergistically. Hydrophobic and amphiphilic α-helices profile. Stable at high temperature and extreme pH. | High efficiency, no drug resistance, have 103 times more active when combined with their complementary peptide than individually and easily amenable through bioengineering to increase either their activity or specificity towards target microorganisms. | Antibiotics candidate, human clinical drug | Bengtsson et al., 2020; Kristiansen et al., 2005; Oppegård et al., 2016; Xu et al., 2019 |
| Class IIc | Small and heat stable peptide, divided into two groups; thiolbiotics and cystibiotics, and N- and C-termini are covalently linked. | It can be used as bacteriocin–receptor complex and employ a similar mechanism to target susceptible cells. | Antibiotics candidate | Ahmad et al., 2017; Collins et al., 2017; Diep et al., 2007 |
| Class IIId | Class IIId is still unclassified bacteriocins, all of the bacteriocins which do not include the criteria of the previous three Class II, including sec-dependent bacteriocins and leaderless bacteriocins. | Heat stable and wide range of pH condition, especially in neutral and alkaline pH | Bread and food industry | Hata et al., 2010; Perez et al., 2014; Todorov, 2009 |
Figure 1 Plantaricin biosynthesis gene cluster *L. plantarum* WCFS1; a – d, *plnW*, *plnV*, *plnU*, and *plnT*, integral membrane protein, respectively; e, *plnS*, two-peptide bacteriocin; f, *plnH*, bacteriocin ABC transporter, accessory factor *plnH*; g, *plnG*, bacteriocin ABC-transporter, ATP-binding and permease protein; h – i, *plnE* and *plnF*, two-peptide bacteriocin; j, *plnD*, response regulator, repressor; k, *plnC*, response regulator, activator; l, *plnB*, histidine protein kinase, sensor protein; m, *plnA*, precursor peptide, induction factor; n, *plnQ*, plantaricin biosynthesis protein; o, *plnP*, bacteriocin immunity protein; p, *plnO*, plantaricin biosynthesis protein; q, *plnN*, bacteriocin precursor peptide; r, *plnM*, bacteriocin immunity protein; s - t, *plnJ* and *plnK*, two-peptide bacteriocin plantaricin JK; u, *plnl*, bacteriocin immunity protein

Many other plantaricins have been reported, such as plantaricin 149 (Kumagai *et al*., 2019), plantaricin JY22 (Lv *et al*., 2018), plantaricin Z15 (Song *et al*., 2014), plantaricin LPL-1 (Wang *et al*., 2018), plantaricin NC8 (Jiang *et al*., 2018), and plantaricin BM-1 (Xie *et al*., 2018). Their different characteristics are influenced by differences in *L. plantarum* strains, the type of genetic location, whether the gene encodes a one- or two-peptide plantaricin, and the amino acid sequence, which affects the interaction with pathogens. Plasmids associated with bacteriocin production vary considerably in size, and plantaricin genes on the plasmid are also found on the chromosome (Wada *et al*., 2009). Some plasmids are known to carry genetic determinants for several plantaricins.

### 4. Inhibitory mechanism of plantaricin

Bacteriocins from different classes differ in their mechanism of bacterial inhibition. Most bacteriocins are cationic peptides (20–70 amino acids) that disrupt the integrity of the cell wall or inhibit protein or nucleic acid synthesis (Ahmad *et al*., 2017). Most class II bacteriocins are synthesized as biologically inactive pre-peptides, carrying an N-terminal leader peptide and a distinctive double-glycine proteolytic processing site. Bacteriocins bind to lipid components of the bacterial cell wall via surface molecular binding sites and facilitate pore formation or direct cell lysis via specific or non-specific receptor binding (Chikindas *et al*., 1993). The binding step involves electrostatic interaction between positive regions of amino acids in the bacteriocin and negatively charged phospholipid groups in the target membrane (Todorov, 2009). Todorov (2009) reported that both N- and C-terminals of class II bacteriocins contribute to membrane binding. The formation of pores may cause the leakage of inorganic phosphates (and subsequent ionic imbalance), β-galactosidase, and genetic materials (DNA/RNA) (Jiang *et al*., 2018; Wiedemann *et al*., 2006). Further, the proton motive force is dissipated by disrupting the transmembrane potential and/or the pH gradient of sensitive cells, followed by cell death (Chikindas *et al*., 1993; Van Reenen *et al*., 2003).

Two-peptide plantaricins such as *plnEF* and *plnJK* appear to form relatively specific pores via dissipating the transmembrane potential, decreasing pH, and inhibiting enzymatic processes (Oppegård *et al*., 2016; Sharma and Srivastava, 2014). Some reports have shown that class II bacteriocins may cause pore formation via the carpet mechanism, whereby peptides orientate themselves parallel to the membrane and interfere with the structure. This may be due to putative transmembrane helices, membrane-binding ability, or solubility in water (Chikindas *et al*., 1993). Some reports show two hypothetical mechanisms of interaction between class II bacteriocins and the membrane surface: electrostatic binding of the bacteriocin to the membrane surface and binding between...
positively charged amino acids and anionic phospholipid heads in the membrane (Drider et al., 2006; Ennahar et al., 2000; Richard et al., 2006), where the N-terminal domain plays a role in the electrostatic interaction between the bacteriocin and the membrane surface and the C-terminal determines cell-specificity (Ennahar et al., 2000; Fimland et al., 1996; Todorov, 2009). The C-terminal hydrophobic/amphiphilic α-helix has a role in the insertion of bacteriocin into the cytoplasmic membrane of target cells, resulting in the formation of water-filled pores, and plays a role in target-cell specificity due to its putative transmembrane helices (Ennahar et al., 2000).

In addition, plantaricin is reported to contain cysteine and aromatic amino acids. The modification of pairs of cysteine residues, to form disulfide bridges, affects antimicrobial activity (Miller et al., 1998), and the loss of cysteines or aromatic amino acids causes a reduction in activity. All of the mechanisms explained above are shown in Fig. 2. In summary, plantaricins have many mechanisms to kill or inhibit bacterial growth, and it is expected that these mechanisms are effective in killing pathogens and could help to extend the shelf life of food. Further, it is beneficial for researches and industry to study the mechanism of applying bacteriocin in diverse industries.
**Table 2 Characterization and Heterologous of Plantaricin**

| Plantaricin | Characterization | Strain | Location | Reference |
|-------------|-----------------|--------|----------|-----------|
| pbnA | Primarily a peptide pheromone that induces bacteriocin production, a member of the non-lantibiotic which encode a precursor protein, small peptides (20-60 amino acids), high isoelectric points, and amphiphilic α-helical peptide | *L. plantarum* LZ206, *L. plantarum* 80, *L. plantarum* C11 | Plasmid and chromosome | Andersland *et al.*, 2010; Kristiansen *et al.*, 2005; Diep *et al.*, 1994 |
| pbnB | A two-component system histidine protein kinase *pbnB* and sensor protein in *L. plantarum* | *L. plantarum* WCFS1, *L. plantarum* NCIMB 700965, *L. plantarum* 10CH, *L. plantarum* 80 | Chromosome | El Halfawy *et al.*, 2017; Heeney and Marco, 2019; Illeghems *et al.*, 2015 |
| pbnC | The locus included structural gene *pbnC* is followed by a gene encoding a LanM-like protein and four downstream genes encoding ABC-type transporter components, mature peptide of 27 amino acid residues, containing one dehydroalanine, one lantionine and, three β-methyl-lanthionine residues, dissipate the proton motive force and to induce release of intracellular molecules such as glutamate and ATP, and belongs to family of lantionine group. | *L. plantarum* LL441, *L. plantarum* 80 | Chromosome | Bruno Bárcena *et al.*, 1998; Flórez and Mayo, 2018; Wiedemann *et al.*, 2006 |
| pbnD | Heat stable, inactivated by α-chymotrypsin, trypsin, pepsin and proteinase K, stable at pH 2–10 | *L. plantarum* BFE 905, *L. plantarum* C11 | Chromosome | (Franz *et al.*, 1998) |
| pbnEF | A two-peptide plantaricin, some reports showed the mechanism was difficult to evade and compared with conventional antibiotics that usually target metabolic enzymes, molecular weight is ~10 KDa included structural bacteriocin, immunity, partial transporter and potential regulatory encoding regions, and amphiphilic α-helices peptide. | *L. plantarum* WCFS1, *L. plantarum* NCIMB 700965, *L. plantarum* 10CH, *L. plantarum* 80 | Chromosome | Anderssen *et al.*, 1998; Heeney *et al.*, 2019; Ghomazadeh *et al.*, 2019; Selegård *et al.*, 2019 |
| pbnG, pbnH | *pbnG* and *pbnH* are bacteriocin ABC-transporter, *pbnG* is ATP-binding and permease protein *pbnG*, and *pbnH* is accessory factor *pbnH* | *L. plantarum* WCFS1, *L. plantarum* NCIMB 700965, *L. plantarum* 10CH, *L. plantarum* 80 | Chromosome | El Halfawy *et al.*, 2017; Heeney and Marco, 2019; Illeghems *et al.*, 2015 |
| pbnI | A bacteriocin immunity protein *pbnI*, membrane-bound protease CAAX family | *L. plantarum* subsp. *plantarum* P8, *L. plantarum* subsp. *plantarum* NC8, *L. plantarum* WCFS1 | Chromosome | Li *et al.*, 2016 |
Table 2 Characterization and Heterologous of Plantaricin (continue)

| Plantaricin | Characterization | Strain | Location | Reference |
|-------------|------------------|--------|----------|-----------|
| plnJK       | A two-peptide of bacteriocin that include in bacteriocin Class Ib, plnJ and plnK. The number of amino acid plnJK is 25 amino acids for plnJ and 32 amino acids for plnK, significant antimicrobial activity toward gram-positive bacteria to inhibit pathogen bacteria to prevent food spoilage, and plnJK is encoded in the plantaricin locus of *L. plantarum* by the genes plnJ and plnK, which are co-transcribed with the immunity genes plnLR, amphiphilic α-helices peptide. | *L. plantarum* LZ206, *L. plantarum* C11 | Chromosome | Ekblad *et al.*, 2017; Li *et al.*, 2016; Oppegaard *et al.*, 2016; Xu *et al.*, 2019 |
| plnL        | Bacteriocin immunity protein | *L. plantarum* LZ206, *L. plantarum* WCFS1, *L. plantarum* ZJ316, *L. plantarum* subsp. plantarum NC8 | Chromosome | Li *et al.*, 2016 |
| plnM, plnN, plnO, plnP, plnQ, plnR | plnM and plnP are encode as immunity proteins. plnO, plnQ, plnR are encoded as biosynthesis protein. These plantaricin are not too much reported, analyzed, and purified, so that difficult to describe its characteristics. | *L. plantarum* C11, *L. plantarum* 80 and *L. plantarum* WCFS1 | Chromosome | Diep *et al.*, 1996; Illeghems *et al.*, 2015 |
| plnS        | A two-peptide of bacteriocin and consists of the 27 residue Pls-α peptide and the 26 residue Pls-β peptide. In plnS, GxxxG and GxxG-like interaction motifs are important for helix-helix interactions in lipid membranes as antimicrobial activity and has aromatic residues such as Trp and Tyr. | *L. plantarum* LPCO10 | Chromosome | Ekblad and Kristiansen, 2019; Jimenez-Diaz *et al.*, 1995; Stephens *et al.*, 1998 |
| plnT, plnU, plnV | plnT, plnU and plnV are integral membrane protein plnU. These plantaricins are not too much reported, analyzed, and purified, so that difficult to describe its characteristics. | *L. plantarum* C11, *L. plantarum* 80 and *L. plantarum* WCFS1, *L. plantarum* subsp. plantarum P-8 | Chromosome | Diep *et al.*, 1996; Illeghems *et al.*, 2015 |
| plnW        | A new two-peptide bacteriocin, inhibits large number of Gram-positive bacteria. *plnW* structural gene sequenced has similarities to two other two-peptide lantibiotics, namely staphylococcin C55 and lacticin 3147, and a member of lantibiotics with common bridging patterns | *L. plantarum* U10 | Plasmid and chromosome | Holo *et al.*, 2001; Lages *et al.*, 2015 |
Table 2 Characterization and Heterologous of Plantaricin (continue)

| Plantaricin | Characterization                                                                 | Strain          | Location  | Reference          |
|-------------|----------------------------------------------------------------------------------|-----------------|-----------|--------------------|
| pbnY        | Strong inhibitory activity against *Listeria monocytogenes* BCRC 14845 and *B. subtilis subsp. subtilis* and heat stable. The molecular mass is 4,296.65 Da | *L. plantarum* 510 | Chromosome | Chen *et al.*, 2014 |
| pbn423      | Small peptide, heat resistance, sensitive to proteolytic enzymes, stable at pH 1–10 | *L. plantarum* 423 | Plasmid   | Van Reenen *et al.*, 2003 |
| pbn LpU4    | Small peptide, has antimicrobial activity, no homology with other bacteriocins, heat stable, sensitive to proteinase K, pepsine and pronase | *L. plantarum* LpU4 | Plasmid   | Milioni *et al.*, 2015 |
| pbn ASM1    | Heat stable, inhibits closed bacterial species, stable in wide pH compared to nisin A | *L. plantarum* A-1 | Plasmid and chromosome | Hata *et al.*, 2010 |
5. Future use of plantaricin

5.1 Bio-preservation of raw meat, seafood and dairy products

*Staphylococcus aureus, Listeria monocytogenes, Bacillus cereus, and Clostridium botulinum* are known bacterial pathogens involved in food safety. Plantaricin is reported to be bactericidal and bacteriostatic for these foodborne pathogens and could be a bio-preservative for marine fish and shellfish products (Čanak et al., 2018), raw turkey meat (Cho et al., 2010), milk (Murua et al., 2013), dairy products (Azizi et al., 2017; Doulgeraki et al., 2013), and fermented foods (Man and Xiang, 2019; Paramithiotis et al., 2019). Plantaricin is nontoxic to humans. It shows heat stability and antibiotic activity and is reported to be easily degraded by proteolytic enzymes because of its proteinaceous nature (Perez et al., 2014). A concentration of plantaricin of up to 5000 mg/kg body weight of mice did not affect the levels of leukocytes, erythrocytes, hematocrit, hemoglobin, platelets, urea, creatinine, alanine transaminase, or aspartate transaminase, and histopathological observation showed a picture of normal liver and kidney cells (Hanny et al., 2019). Although the use of plantaricin is safe for humans, it has not been effectively used as a bio-preservation in the industry because one of its limitations is that it can kill and inhibit closely related species. There are no studies of the use of synthetic plantaricin in large industry.

5.2 Bio-preservation of raw plant material

Bacteriocin has been tested as part of an edible coating that may reduce microbial growth and decay in perishable fruits, thus improving product shelf life and commercial appearance (Tumbarski et al., 2019). Bacteriocin enriched with 0.5% carboxymethyl cellulose was reported to be effective in the extension of the shelf-life of fresh strawberries (Tumbarski et al., 2019). Partial purification of plantaricin Gt2 from *L. plantarum* UTNGt2 was reported to disrupt the membrane of *E. coli*, causing the release of β-galactosidase and leakage of DNA/RNA molecules, followed by cell death, revealing a bacteriolytic mode of action. Plantaricin Gt2 is similar to plantaricin W, stable at pH 2–10, and is active in killing *Salmonella* and *E. coli* in the human body (Tenea and Pozo, 2019). Tomato fruits coated with plantaricin Gt2 showed growth inhibition of an artificially inoculated *Salmonella* cocktail, demonstrating its potential as a preservative (Tenea and Pozo, 2019). This coating technology, which has yet to be adopted by the industry, could extend the shelf life of fruits and vegetables by killing spoilage bacteria. This will help extend the shelf life and market fruits without harmful preservatives so that these benefits can be felt by all, without danger to health. These plantaricins can be used as a coating product based on the diverse structures and activities, but unfortunately, this technology has not yet been developed by industry. This could also lead to the development of novel plastic containers that can store food for longer.

5.3 Antibacterial packaging

Plantaricin was developed as an active antibacterial packaging film by coating a polyethylene terephthalate/polyvinylidene chloride/retort casting polypropylene multilayer film with plantaricin BM-1 and chitosan, for chilled meat preservation (Yang et al., 2019). This was reported to prolong the shelf life of pork during cold storage (Xie et al., 2018). The active plantaricin film significantly decreased the surface population of *L. monocytogenes*, compared to the control (Yang et al., 2019), and is reported to cause direct cell disruption (Ercolini et al., 2006). Plantaricin is also reported to control *L. monocytogenes* and spoilage bacteria in sliced, vacuum-packed ham, without the addition of chemical preservatives (including sodium nitrite) over 35 days of storage at 4 °C (Zhang et al., 2017; Zhou et al., 2015). The development of this packaging will make food more durable, as it can kill surface pathogens, but has yet to be applied as a method by the food industry. This may be due to reports that plantaricin kills other, non-pathogenic, closely related species (Malik et al., 2016). Collected
data show that plantaricin is effective against diverse bacteria, and also against eukaryotic microbes such as *Candida albicans* (Sharma and Srivastava, 2014). Plantaricin has a lower activity than antibiotics, although it maintains its activity at high temperatures and in acidic and saline conditions. This technology is expected to develop in the future, in line with the movement for more natural foods, with fewer chemical additives.

### 5.4 Anti-*Candida*

Plantaricin has been reported as an inhibitor of *C. albicans*. Plantaricin plnJ was identified as more effective than plnE, plnF, and plnK. PlnJ not only induced dissipation of membrane potential but also resulted in the release of K⁺. Treatment with plantaricins initiates apoptotic cell death, which may lead to necrosis due to toxicity of the plantaricin peptides (Sharma and Srivastava, 2014). This knowledge will direct researchers to develop anti-*Candida* biofilms that are beneficial to humans.

### 5.5 Anti-cancer

Plantaricin P1053 isolated from *L. plantarum* PBS067 strain is one of the first multifunctional bacteriocin-like compounds to be found to act on human epithelial intestinal cells. The molecular mass of plantaricin P1053 is 1053 Da (De Giani *et al*., 2019). Plantaricin P1053 showed normal effect and cancerogenic epithelial intestinal cell lines through an enhancing of viability of healthy cells and a proliferation reduction of cancer cells. Little research has been done on plantaricin as an anti-cancer agent, but this is a promising area for investigation.

### 5.6 Cholesterol-lowering and antioxidant activity

PlnE and plnF are reported to improve leukocyte, hematocrit, and hemoglobin levels as well as decrease malondialdehyde levels (Hanny *et al*., 2019). Fourteen different plantaricin-encoding genes at pln loci in the *L. plantarum* strain were reported to possess cholesterol-lowering and antioxidant activity (Devi and Halami, 2019). These benefits could be applied directly in the development of functional foods.

### 6. Conclusion

Plantaricins are varied and have different characteristics and mechanisms to protect their host cells from unfavorable environments. *L. plantarum* is a lactic acid bacterium that produces plantaricin to kill and inhibit bacterial pathogens, by various mechanisms. Plantaricin produced from each *L. plantarum* strain is encoded on plasmids, chromosomes, and transposons. The many functions and applications of plantaricin are of great interest to the food industry and should provoke future innovation.

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