Transactions on Materials, Biotechnology and Life Sciences
ISSN: 2960-1932, eISSN: 2960-2319 | Volume 3
BLSME 2024

Nisin—A Promising Antimicrobial Compound from Lactic Acid Bacteria

Guanchong Chen

College of Life Science, South China Agriculture University, Guangzhou, 510000, China

Abstract. Nisin is a classical bacteriocin consisting of thirty-four amino acid residues and undergoing a high degree of post-translational modification to form a unique pentacyclic structure. Nisin is mainly synthesised by lactic acid bacteria. Under natural conditions, it exhibits strong antibacterial activity against Gram-positive organisms and, when combined with chelating agents, is effective against Gram-negative organisms. An excellent safety profile, low cytotoxicity and slow development of resistance are additional benefits. Currently, the application of nisin is focused on food preservation. This is due to the fact that nisin is naturally present in a wide range of historical fermented foods and its safety is fully guaranteed. With the continuous development of bacterial drug resistance and the consequent demand for new anti-infective drugs, its biomedical applications are being actively explored. However, due to the problems of drug stability, safety, and in vivo activity, there is still a certain distance from clinical application. In this paper, the author summarized the structure, biosynthetic mechanism, applications, and possible research directions of nisin.

Keywords: Nisin; Gram-positive organisms; bacterial drug resistance; anti-infective drugs.

1. Introduction

Nisin is an ancient antimicrobial compound that was discovered in 1928 the same year as the world's first antibiotic, penicillin. At the time, people discovered that lactic acid bacteria had an inhibitory effect on starter cheese cultures [1]. This peptide was named nisin twenty years later. Nisin is a peptide with 34 amino acid residues. Unique post-translational modifications give it some unusual amino acid residues, which are lanthionine (Lan) and methyllanthionine (MeLan). These amino acid residues form five lanthione bridges, and these special structures are the basis of nisin's action [2]. In nature, nisin can be synthesised from different L. lactis strains.

Nisin inhibits Gram-positive (GP) bacteria across a wide range of pathogens. Nisin also effectively inhibits Gram-negative (GN) bacteria when combined with chelating chemicals like EDTA [3].

Nisin's current applications are focused on food preservation. It has excellent antimicrobial properties and because it is derived from Lactobacillus, a strain often used in food production, there is great confidence in its oral safety [4]. In recent years, due to the development of bacterial resistance, there has been an urgent search for alternatives to existing antibiotics in order to prevent us from falling into a "post-antibiotic era" where no drugs are available. Nisin, a bacteriocin that inhibits a wide range of pathogenic bacteria and is less likely to develop resistance, is definitely one of the areas that need to be explored. Currently, the application of nisin in medicine is under constant research and encouraging results have been achieved [5]. However, it is important to note that nisin is still a long way from being used in large-scale medical applications.

In this paper, the author summarized the structure, biosynthetic mechanism, and applications of nisin. And at the end, they point out the possible research directions.

2. The structure of nisin

Nisin is a class of genetically encoded host defence peptides (HDP), also known as antimicrobial peptides (AMP). It has a variety of variations, including nisin A, nisin F, nisin U, nisin H, nisin Q, nisin Z, etc. Among them, nisin A is the earliest discovered nisin [2]. The difference between different nisin variants is mainly the substitution of some amino acid residues. For example, nisin Z, the nisin

variant most similar to nisin A, is distinct from nisin A in the amino acid residue at position 27. In nisin A, this is a histidine, whereas in nisin Z, this is an asparagine. [6]. Nisin Q, a nisin extracted in Lactococcus lactis 61-14, which is distinct from nisin A due to amino acid variations at positions 15, 21, 27, and 30 [7]. Among the numerous nisins, nisin A is the most well-studied one. The author will use nisin A to demonstrate the structure of nisin in this paper.

Nisin A is a protein made up of 34 amino acid residues. It contains five lanthione bridges, which help maintain its conformation, one of the keys to its antimicrobial action. Nisin also contains three amino acid residues not commonly found in other proteins, which are dehydrobutyrine and β-methyl-lanthionine, dehydroalanine. Depending on the position and function, nisin can be divided into a C-terminal domain and an N-terminal domain, which will play different roles in the bactericidal process. Figure 1 shows its structure and partitioning. The N-terminal structural domain of nisin has a high affinity for lipid II on the bacterial cell wall. And it forms intermolecular hydrogen bonds lipid II and eventually changes into a hollow cage-like structure, which contributes to the subsequent disruption of the bacterial cell membrane [8]. Furthermore, according to Peter 't Hart and his associates, this binding process is dependent on the pyrophosphate group on lipid II. If the pyrophosphate portion of lipid II is removed, the affinity of nisin for lipid II decreases dramatically [9]. After the N-terminal sequence binds to the cell wall and forms a cage-like structure, the C- terminal sequence of nisin flips over into the bacterial cell membrane [10]. In this process, the hinge-like region that connects the two structural domains also plays an important role.

It is currently believed that because of the small relative molecular mass of nisin, multiple nisin molecules need to be involved in the disruption of bacterial cell membranes to form pore complexes. The formation of the pore complex was shown to be a two-step process by Hester Emilie Hasper et al. A total of 8 nisin molecules with 4 lipid II molecules are required for this process. The first step is to form a cage-like structure by binding the four nisin molecules to the lipid II molecules in a 1:1 ratio. The pore complex will thereafter be formed by the recruitment of four more nisin molecules [11].

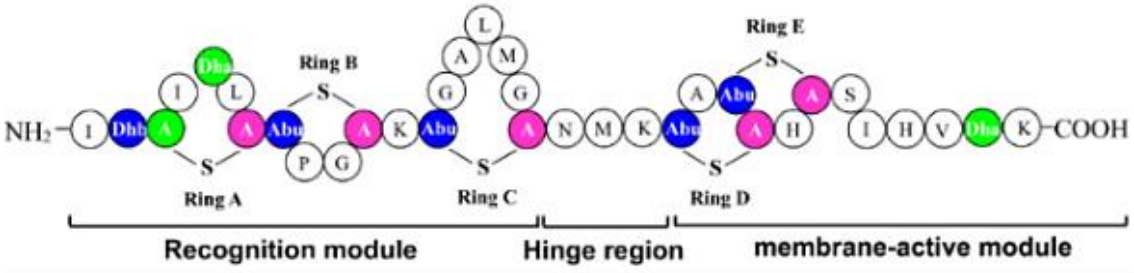

Figure 1. Structure of nisin peptide chain and its partitioning [2].

3. Nisin biosynthesis and its regulation

Nisin is synthesised from an encoded precursor peptide termed NisA, just like the majority of bacteriocins. The core peptide (CP), which is placed at the C-terminal end of nisin A, and the leader peptide (LP), which is located at the N-terminal end, make up nisin A. The LP aids in the recognition of NisA during modification and transport and is removed in the final product. The CP undergoes post-translational modification and is retained in the final product, where it acts as an antimicrobial agent. NisA undergoes post-translational modification in three major steps [2].

The first step is the dehydration of tryptophan and serine. After this step, some of the amino acids in NisA are converted to dehydroalanine (Dha, from serine) and dehydrobutryine (Dhb, from tryptophan). This process is completed in the enzyme dehydratase called nisB. NisB dehydrates NisA a total of eight times, which dehydrates three Ser and five Thr in the CP. Recent studies have shown that this process is accomplished by a glutamylation mechanism, whereby glutamate is transferred to the serine and tryptophan side chains in the CP, and then serine and tryptophan are eliminated to produce dehydroalanine and dehydrobutynine. [12].

The second step in Nisin biosynthesis is the cyclisation of dehydrated residues. In this process, the amino acids that were dehydrated in the first step are linked to cysteine to form five rings. They are a lanthionine (formed by linking threonine to cysteine) and four methyllanthionines (formed by linking tryptophan to cysteine). Olli Koponen et al. demonstrated that this process occurs with the cyclase nisC by using a mutant Streptococcus lactis strain without nisC. They introduced His-tagged nisin precursor peptides into Streptococcus lactis mutants lacking nisB or nisC, respectively. The results were that the nisin precursor peptide of the mutant lacking nisB was completely unmodified, whereas the nisin precursor peptide of the mutant containing nisB but lacking nisC was dehydrated but no lanthionine was formed. This indicates that the role of nisC in nisin biosynthesis is to cyclise dehydrated residues and form lanthionine [13].

The LP is removed as the last step in the biosynthesis of nisin. Nisin will be transported out of the cell by the specific ABC-type transporter NisT before the LP is removed. [14]. Outside the cell, the serine protease NisP will remove the LP. Studies have shown that nisin will not possess biological activity if the LP is not correctly removed. The author speculate that this mechanism may be mechanism may be to protect the secreting cells from nisin secreted by itself. Due to the fact that two immune systems—the lipoprotein NisI and the ABC transporter protein NisFEG—are exclusive to secretory cell membrane surfaces. They are able to shield the cell membrane. There's no evidence to support that this immune system exists inside the cell membrane. [15]. Therefore, secretory cells may need to ensure that nisin is inactive before it is expelled from the cell membrane in order to avoid the breakdown of the cell membrane from the inside by nisin, which can lead to cell death. In summarized form, Figure 2 lists every post-translational modification process of Nisin.

Several biomolecules in the cell are in charge of controlling nisin production in addition to the proteins that are directly involved in it. A two-component signal transduction system is made up of NisK and NisR. Mature nisin attaches to and activates NisK, which then transfers the phosphate group to activate NisR, so controlling nisin gene production [16]. A more comprehensive summary of the function and co-operation of the various proteins involved in nisin biosynthesis is given by the Fig.3.

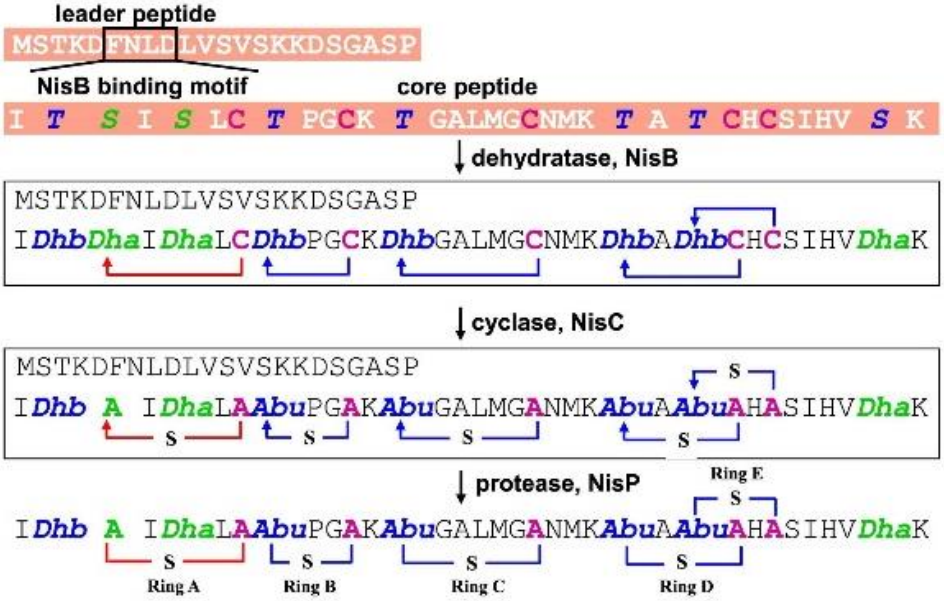

Figure 2. Post-translational modification process of Nisin [2].

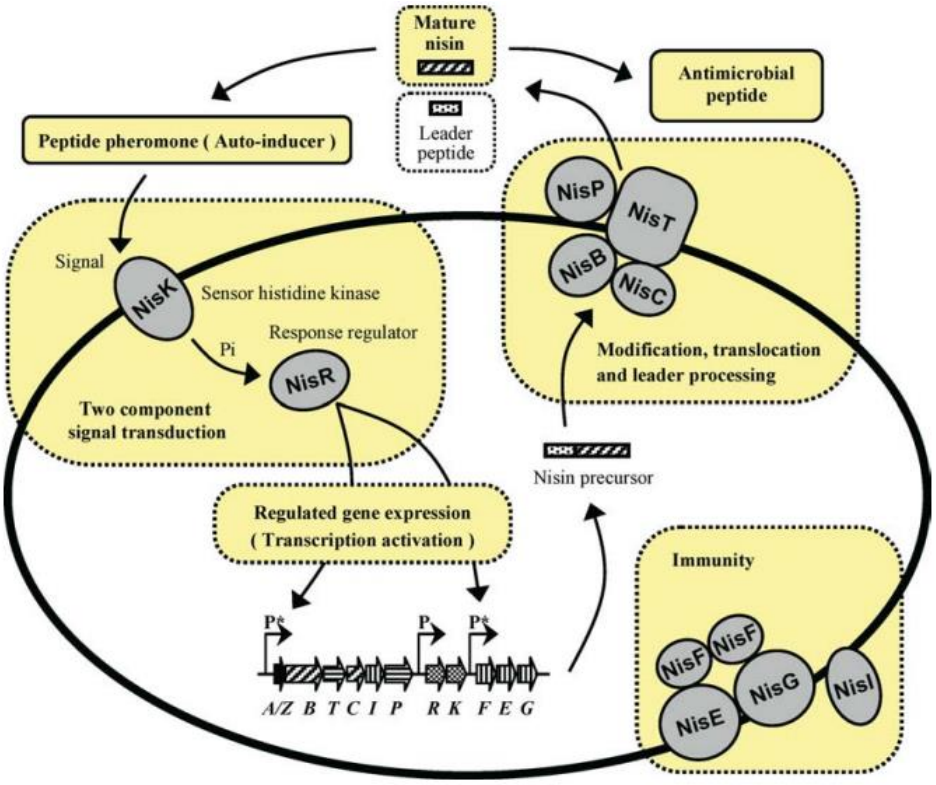

Figure 3. Schematic representation of nisin biosynthesis and regulation [2].

4. The applications of nisin

Ever since nisin was discovered by Rogers and Whittier in 1928 for its inhibitory effect on fermented cheese cultures, research has been conducted on nisin [1]. There is certainly no lack of exploration of its applications, including food preservation, anti-infective, anti-cancer and so on. But due to some characteristics inherent in nisin as a peptide, such as low absorption rate and sensitivity to environmental changes such as PH, its application has not been promoted as well as penicillin, which was discovered at the same time. However, at present, with the development of genetic engineering, synthetic biology, bioinformatics and other technologies, the application of nisin, especially in the direction of biomedicine, has rekindled people's interest. In the following sections, the author will explore the applications of nisin in a number of different areas.

4.1. Food preservation

The food preservation aspect is the most mature application of nisin compared to other aspects. Thanks to the fact that it is derived from Streptococcus lactis, a bacterium widely used in the production of a variety of food products, it quickly gained acceptance for its safety. At the moment, nisin is the sole bacteriocin that can be used in food. The use of nisin in food is permissible by regulatory authorities in more than 80 countries, including the FDA in the United States and EFSA in the European Union. Its applications include dairy products, cereals, meats, fruit juices, vegetables and more. In fact, nisin is active against many food-destroying and disease-causing microorganisms, such as Listeria, Staphylococcus, Clostridium, and Bacillus spp [4].

The use of nisin in the preservation of the foods has a number of advantages, such as prolonging the shelf life of foods, cutting back on the usage of chemical-based preservatives, and lowering the amount of salt used in some foods as a preservative to make them healthier. When used alone, nisin is lethal to GP bacteria, while it is not as effective against GN bacteria. This is due to the fact that the surface of the GN bacteria is encapsulated by layers of lipopolysaccharides, which prevent nisin from reaching the cell wall of the GN bacteria and thus prevent it from working. [4]. Many common food pathogenic bacteria such as Salmonella are also GN, which makes nisin insufficient for food

preservation when used alone. To solve this problem, nisin can be used together with the chelating agent EDTA. According to Z. R. Liang et al. a mixture of low concentrations of nisin, EDTA and sulphite was effective in reducing the number of viable bacteria during ice storage of fresh white shrimp [3]. This is because the chelating agent can dissolve the stability of the lipopolysaccharide layer on the surface of GN bacteria, allowing nisin to be transported in the lipopolysaccharide layer and ultimately act with the cell membrane [17].

In order to overcome the previously mentioned disadvantages of nisin, such as its environmental sensitivity, and to reduce the effect of nisin on the flavour of the foodstuff, nisin can also be incorporated into a polymer film used as a foodstuff package when it is used as a preservative. When incorporated into a polymer film, the activity of nisin is better maintained than when it is added directly to food, and the concentration does not decrease due to diffusion into the food. This technique has been tried with satisfactory results in sliced sausages and mangoes [18, 19].

4.2. Anti-infective

Although there are now well-established therapeutic options for anti-infection treatment, namely antibiotics, infections due to multi-drug resistant organisms (MDROs) are increasing with the development of bacterial resistance. Hospitals worldwide are facing a significant issue with MDRO-caused infections, including vancomycin-resistant enterococci (VRE) and methicillin-resistant Staphylococcus aureus (MRSA). Diekema et al. report that the detection rate of MRSA in instances of S. aureus infections was 26.3% in Europe, 46% in the Western Pacific area, 34.9% in the United States of America, and 46% in Latin America. [20]. Typically, infections due to MDRO occur in hospital settings. However, in recent years, community-associated MDRO infections have also begun to increase. For example, the community-associated MRSA. They have the power to infect healthy populations outside of hospitals and possess both methicillin resistance and enhanced virulence, threatening public health [21]. In summary, there is a need for a novel anti-infective treatment regimen to address the growing problem of bacterial drug resistance. And the food industry has been using nisin to inhibit the growth of pathogenic bacteria for decades with an excellent safety record. This has inspired us to apply nisin in the anti-infective field to combat MDRO infections.

There have been many studies on the use of nisin as an anti-infective drug, especially with regard to the currently intractable problem of MDRO infections. The present study findings indicate that nisin inhibits a variety of infections, including those of the gastrointestinal tract, respiratory system, skin and soft tissue, mastitis, mouth infections, etc. However, the relevant tests are stuck in in vitro experiments or animal experiments, which need to be further investigated [5]. When treating infections caused by drug-resistant bacteria, Clare Piper et al. concluded that nisin has a better effect in inhibiting MRSA, with MICs ranging from 0.5-4.1 mg/L against many different MRSA strains. They believe that nisin can be combined with other antimicrobial compounds in future studies and become clinically used antimicrobial agents [22]. Leonides Fernández et al. showed that nisin could have a therapeutic role in mastitis. They applied nisin solution to the areola of women with clinical signs of staphylococcal mastitis. After 14 days, the breast milk of women who had received nisin treatment had far fewer staphylococcal counts than that of the control group. Over the course of 14 days, the women who administered the nisin solution did not exhibit any clinical symptoms of mastitis, while the women in the control group continued to show signs of mastitis throughout the study. And because nisin is widely permitted to be used as a peptide in food, its effect on the foetus was negligible. They concluded that nisin appears to be a successful substitute for antibiotics in the management of staphylococcal mastitis [23].

Besides treating infectious diseases, it is also thought to be useful for addition to macromolecular polymer membranes or fibres to inhibit microbial growth and biofilm formation. This has positive implications for avoiding infections caused by bacteria adhering to medical instruments or other surfaces and warrants further investigation [24, 25].

4.3. Anti-cancer

As an AMP, much of the research on nisin has focused on its effects on microorganisms. However, some recent studies have also focused on its anticancer effects and concluded that nisin has the potential to control the proliferation of cancer cells. The explanations for its anticancer mechanism can generally be divided into three categories. Firstly, nisin can inhibit the increase of cancer cells by specifically inducing apoptosis. E Joo et al. found that nisin inhibited head and neck squamous cell carcinoma (HNSCC) by treating mice with oral cancer and cancer cell lines with nisin. After nisin treatment, there was a significant increase in cellular DNA fragmentation in the HNSCC cell line, whereas primary oral keratinocytes did not exhibit such characteristics. They found that nisin treatment upregulated the expression of CHAC1, a cation transporter that promotes the transport of apoptotic cations to induce apoptosis [26]. This phenomenon might arise from variations in the composition, function, and reactivity to calcium flow of lipid membranes between primary oral keratinocytes and HNSCC cells [5].

The second mechanism is due to the fact that nisin can slow down the proliferation of cancer cells by reducing the expression of cyclins. It has been found that nisin can down-regulate the expression of cancer-related proteins, including cyclin D1 and the transcription factor TWIST1 [27, 28]. Nisin also binds to the FZD7 protein and inhibits its action [28]. The third scenario is based on the fact that some microbial infections may play a role in inducing cancer. Nisin can fight cancer by treating related microbial infections, such as the well-known gastric H. pylori infection [5, 29]. In this type of case, nisin is certainly a good choice, as it can act both against infection and cancer at the same time.

5. Future direction

When looking at future directions for nisin research, the author believe that two key areas are particularly important: the screening of innovations in nisin production methods and the bioengineering of nisin for improvement. This is not merely a matter of scientific interest but is directly linked to how to tackle the major challenges facing society today. As a biomolecule, nisin is difficult to synthesise directly via chemistry, so it still relies on biosynthesis. However, the yield of nisin by conventional fermentation methods is low [30]. Nisin's future applications in food preservation and biomedicine require better affordability of nisin. Innovative nisin production methods are expected to improve the production efficiency and reduce the production cost of nisin, thus meeting the increasing demand in the food and biomedical markets. By bioengineering nisin to improve it, scientists can better tailor its properties for better therapeutic efficacy or to cope with emerging drug-resistant strains and diseases. The author believe that these two directions will be the focus of Nisin research in the future, as they are directly related to real problems in the fields of food science, medicine and the environment.

In terms of innovative nisin production methods, current nisin production methods mainly utilise natural strains fermented in milk or whey. The production process consists of many unfavourable factors for the strains. They include the inhibition of strains by excessive substrate or lactic acid concentration, the counteracting effect of excessive nisin concentration on strains, and so on. Among them, the counteracting effect of nisin on the production strains cannot be ignored. Even though nisin-producing strains possess an immune system against nisin, i.e., the nisFEG complex, nisin-producing strains are still inhibited by nisin in high nisin concentrations. This is one of the reasons for lower nisin production under traditional methods. The recent successful recombinant production of nisin in Corynebacterium glutamicum using a two-step method by Dominik Weixler et al. provides ideas to address this inhibition [31]. They first introduced into Corynebacterium glutamicum a plasmid containing the genes for nisZ, nisB, nisT, and nisC, which are required for the biosynthesis of Nisin Z. They succeeded in expressing the inactive nisin precursor without excision of the LP in Corynebacterium glutamicum. Subsequently, the active nisin was obtained by treating the obtained prenisin with soluble nisP synthesised in E. coli. nisin synthesis using this method avoids the inhibitory effect of nisin on the production strain. Although this method currently synthesises nisin

at an efficiency similar to conventional commercial synthesis, this is based on laboratory conditions that have not been further optimised [31]. It is believed that methods such as batch replenishment and continuous fermentation, which are common in the modern fermentation industry, can further increase the yield of this production process. Given that the method of stepwise synthesis can completely avoid the feedback inhibition of nisin to its producer that is difficult to avoid in other productions, the author believe that this method is very informative and deserves further study.

Bioengineering can help us to change some amino acid residues of nisin or add other sequences to the c-terminus. The structural sections of nisin are N-terminal, hinge, and C-terminal, and mutagenesis of amino acid residues in different regions will yield different results. By adjusting the amino acid participation of nisin, scientists can enhance its activity against specific species of pathogenic microorganisms. Or change its physicochemical properties, such as solubility and stability under different ph. These modifications have the potential to meet the diverse needs of society in food, biomedicine, environment, and other fields, thereby broadening the range of applications for nisin. As mentioned earlier, the limited application of nisin is partly due to its low solubility and sensitivity to the environment, among other things. Bioengineering is the answer to this problem. For example, the derivatives of Nisin Z, N20K and M21K, have higher high-temperature stability, expanding its application in the food industry. There are also attempts to increase the stability of nisin in the gastrointestinal environment by adjusting the enzyme cleavage site, which, if successful, would be of great significance for the biomedical application of nisin.

6. Conclusion

As bacterial resistance continues to develop, the need for novel anti-infective drugs is increasing. AMP are one of the most promising areas for the birth of novel antimicrobial drugs. This is due to the fact that AMP has a unique site of action, lower cytotoxicity, and lower propensity for drug resistance. In many organisms that do not have a specific immune system, AMP is their primary means of fighting infections with good results. Predictably, if AMP can be successfully applied to anti-infection therapy, the benefits are enormous. And nisin could be an attempt for this application. As one of the most well-studied AMPs, nisin has advantages that other AMPs do not have: clear structure, widely recognised safety profile, excellent bacteriostatic effect, untapped bioengineering potential... These properties make nisin a promising candidate for novel anti-infective drugs. Although the application of nisin is still focused on the field of food preservation. However, nisin research has progressed to the point where it is being considered as a veterinary antimicrobial drug, and it is reasonable to believe that nisin will be used on a larger scale in the medical field in the near future. Considering the uniqueness of nisin's site of action, its excellent bioengineering applicability, and its low cytotoxicity, the author believes that nisin will become an even better anti-infective solution than existing antibiotics.

Nisin also has many other application directions, such as the manufacture of antimicrobial materials, cancer therapy etc. It is a peptide with multi-faceted potential. Thanks to the increasing enthusiasm for research in recent years, the author believes that nisin's great potential will be uniquely realised in this century.

References

[1] A. D. P. van Staden, W. F. van Zyl, M. Trindade, L. M. T. Dicks, C. Smith, Appl. Environ. Microbiol., 87 (14), e0018621 (2021).

[2] J. Wu, M. Zang, S. Wang, B. Zhao, J. Bai, C. Xu, Y. Shi, X. Qiao, Food Microbiol., 111, 104207 (2023).

[3] Z. R. Liang, H. I. Hsiao, D. J. Jhang, J. Food Saf., 40 (4), 12794 (2020).

[4] S. Khelissa, N. E. Chihib, A. Gharsallaoui, Arch. Microbiol., 203 (2), 465 - 480 (2021).

[5] J. M. Shin, J. W. Gwak, P. Kamarajan, J. C. Fenno, A. H. Rickard, Y. L. Kapila, J Appl. Microbiol., 120 (6), 1449 - 65 (2016).

[6] N. Schneider, K. Werkmeister, M. Pischetsrieder, Food Chem., 127 (2), 847 - 54 (2011).

[7] T. Zendo, M. Fukao, K. Ueda, T. Higuchi, J. Nakayama, K. Sonomoto, Biosci. Biotechnol. Biochem., 67 (7), 1616 - 9 (2003).

[8] S. T. Hsu, E. Breukink, E. Tischenko, M. A. Lutters, B. de Kruijff, R. Kaptein, A. M. Bonvin, N. A. van Nuland, Nat. Struct. Mol. Biol., 11 (10), 963 - 7 (2004).

[9] P. t Hart, S. F. Oppedijk, E. Breukink, N. I. Martin, Biochemistry, 55 (1), 232 - 7 (2016).

[10] M. Musiejuk, P. Kafarski, Pharmaceuticals (Basel), 16 (8), 1058 (2023).

[11] H. E. Hasper, B. de Kruijff, E. Breukink, Biochemistry, 43 (36), 11567 - 75 (2004).

[12] N. Garg, L. M. Salazar-Ocampo, W. A. van der Donk, Proc. Natl. Acad. Sci. U.S.A., 110 (18), 7258 - 63 (2013).

[13] O. Koponen, M. Tolonen, M. Qiao, G. Wahlström, J. Helin, P. E. J. Saris, Microbiology (Reading), 148 (Pt 11), 3561 - 3568 (2002).

[14] C. I. Cheigh, Y. R. Pyun, Biotechnol. Lett., 27 (21), 1641 - 8 (2005).

[15] D. Field, M. Fernandez de Ullivarri, R. P. Ross, C. Hill, FEMS Microbiol Rev., 47 (3), fuad023 (2023).

[16] M. Kleerebezem, Peptides, 25 (9), 1405 - 14 (2004).

[17] R. Pattanayaiying, H. K. A, C. N. Cutter, Int. J. Food. Microbiol. 188, 135 - 46 (2014).

[18] B. Marcos, T. Aymerich, M. Garriga, J. Arnau, Food Control, 30 (1), 325 - 330 (2013).

[19] A. A. Barbosa, H. G. Silva de Araújo, P. N. Matos, M. A. Carnelossi, A. Almeida de Castro, Int. J. Food. Microbiol., 164 (2-3), 135 - 40 (2013).

[20] D. J. Diekema, M. A. Pfaller, F. J. Schmitz, J. Smayevsky, J. Bell, R. N. Jones, M. Beach, Clin. Infect. Dis., 32 Suppl 2, S114 - 32 (2001).

[21] M. Otto, Int. J. Med. Microbiol., 303 (6-7), 324 - 30 (2013).

[22] C. Piper, L. A. Draper, P. D. Cotter, R. P. Ross, C. Hill, J. Antimicrob. Chemother., 64 (3), 546 - 51 (2009).

[23] L. Fernández, S. Delgado, H. Herrero, A. Maldonado, J. M. Rodríguez, J. Hum. Lact., 24(3), 311 - 6 (2008).

[24] K. Okuda, T. Zendo, S. Sugimoto, T. Iwase, A. Tajima, S. Yamada, K. Sonomoto, Y. Mizunoe, Antimicrob. Agents Chemother., 57 (11), 5572 - 9 (2013).

[25] J. J. Ahire, L. M. Dicks, Probiotics Antimicrob. Proteins, 7 (1), 52 - 9 (2015).

[26] N. E. Joo, K. Ritchie, P. Kamarajan, D. Miao, Y. L. Kapila, Cancer Med., 1 (3), 295 - 305 (2012).

[27] S. S. Hosseini, H. Goudarzi, Z. Ghalavand, B. Hajikhani, Z. Rafeieiatani, M. Hakemi-Vala, Iran. J. Microbiol., 12 (5), 424 - 430 (2020).

[28] P. Balcik-Ercin, B. Sever, Chem. Biol. Interact., 366, 110152 (2022).

[29] T. S. Kim, J. W. Hur, M. A. Yu, C. I. Cheigh, K. N. Kim, J. K. Hwang, Y. R. Pyun, J. Food. Prot., 66 (1), 3 - 12 (2003).

[30] B. Özel, Ö. Şimşek, M. Akçelik, P. E. J. Saris, Appl. Microbiol. Biotechnol., 102 (15), 6299 - 6307 (2018).

[31] D. Weixler, M. Berghoff, K. V. Ovchinnikov, S. Reich, O. Goldbeck, G. M. Seibold, C. Wittmann, N. S. Bar, B. J. Eikmanns, D. B. Diep, C. U. Riedel, Microb. Cell Fact., 21 (1), 11 (2022).