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Pros of Lactic Acid Bacteria in Microbiology: A Review

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Abstract

The application of Lactic acid bacteria (LAB) crossways many industry segments has consistently steered to both financial and environmental paybacks comprising less affluent processing, improved product quality, completely new products, and biologically sustainable dispensation relation to conventional operations. Lactic acid bacteria produce abundant bactericidal proteins in dairy foods. It is because of the release of organic acids as well as the production of other ingredients such as reuterin ethanol, diacetyl, H_2O_2, and bacteriocin. Many bacteriophages with industrial perspective are purified and characterized. The use of starter cultures producing bacteriocin in fermented foods has been studied in laboratory fermentation processes *in vitro* as well as *in vivo* on a pilot scale. The encouraging results of these studies demonstrated the important role of lactic acid bacteria in the food industry as a starter culture to improve the quality and safety of foods. Thus, this review summarized some of the most important services of LAB and the massive amount of research going on related to every aspect for the maintenance of human beings plus humanoid health in terms of healthy and nutritious foods.

1. Introduction

Lactic acid bacteria (LAB) that are generally considered to be safe and useful, are mostly the Gram-positive bacteria including lactobacillus, Lactococcus, and Streptococcus. The coordination is calculated to account for bacterial and gram positive aggregation, the non-spore formation of coconuts or ribbons, which are capable to produce lactic acid as a main product during the fermentation process of carbohydrates. LAB and traders consume vitamins, amino acids (a.a), carbohydrates nucleic acid and peptides. The current taxonomic reviews of these races indicated that lactic acid bacteria include: salivary, Corynebacterium, intestinal, Lactococcus, Clostridium and Streptococcus [1].

Lactic acid bacteria are fastidious in nature, require anaerobic conditions to grow, , lack cytochromes, strictly fermentative and acid-tolerant. They're typically non-motile and non-sporulating microorganisms which produce lactic acid. The clusters of these microorganisms include both rod shaped bacteria and cocci. Different species of LAB have modified to raise underneath various harsh condition from low to high temperature. Their common habitats are the alimentary tract of different animals, dairy products, food products, and soil. Though LAB are not dominant within the traditional enteric microbiota, many trials are commenced to induce a man-made dominance of lactic acid bacteria [2].

Lactic acid bacteria are classified in totally diverse species with adopted morphological form, glucose fermentation pattern, progression at completely altered temperatures, the formation of Lactate and their capability to nurture at high concentrations of salt, acid and alkaline tolerance. LAB are principally distributed into 2 groups based upon the end-products of fermentation of aldohexose. Homofermentative LAB such as Pediococcus, Streptococcus, Lactococcus, and a few Lactobacilli which produce carboxylic acid as the only end-product of aldohexose fermentation. Heterofermentative Lab, i.e. Leuconostoc and Weissella and some of the Lactobacilli as they yielded the equimolar amount of CO_2, alcohol, and lactate from glucose through the monosaccharide monophosphate or simple sugar pathway [3].

2. Lactic acid bacteria as a Probiotics

The term "probiotic" originates from the Greek word “life” Greek word "life" and is currently being used to name the associated bacteria which are beneficial to humans and animals. According to the World Health Organization (WHO) guidelines, probiotics are known as the living organisms which on administration in sufficient quantities, bring health benefits to the host. Probiotics were first investigated in 1908 by Elie Metchnikoff, a medical Nobleman. He suggested that proteolytic microbes in the colon produce toxic substances responsible for the aging process and proposed that consuming fermented milk causes LAB coatings in the colon, lowers intestinal pH, inhibits proteolytic bacteria, and slows down the aging process [4].

In order to qualify for probiotics, the bacteria need to meet certain criteria: the bacterial strain must be fully identified, ingested harmlessly, adhered to the mucosa, capable of colonizing the intestinal epithelium, stable during storage, and must persist in the acid to survive the concentration of bile salts of gastrointestinal tract [5]. Researchers use probiotics under a variety of medical conditions. Bowe & Logan, [6] discussed the prospects of probiotics for the treatment of acne vulgaris, although there have been no suitable trials to date. Probiotics can also be used to avert intestinal obstruction dysfunction in acute pancreatitis [7].

Probiotic pre-treatment weakened acute pancreatitis influenced proliferation in *E. coli* passage [Probiotics 57.4 vs. placebo 223], Cr-EDTA flux [16.7 vs. 32.1 cm/s 10-6], apoptosis and lipid peroxidation [0.42 vs. 1.62 pmol MDA/mg protein]. Ouwehand *et al.*, [8] reported the efficacy of *Lactobacillus acidophilus* “North Carolina Food Microbiology” in the alleviation of allergic rhinitis. Probiotic strain *Lactobacillus fermentum* VR1-033PCC reduced atopic dermatitis in 56 children with age between 6-18 months and reduced the cases by 54% as compared to only 30% in the placebo group. Probiotic bacteria were also studied for their useful effect on autoimmune encephalomyelitis [09], childhood constipation [10], hypertension [11] and found effective.

3. Production of Bacteriocins by LAB

Bacteriocin is a naturally active protein molecule using bactericidal mode of action. Bacteriocin can act as an anti-competitive agent that allows strains to invade established microbial communities. Bacteriocins produced by Gram +ve bacteria are diverse and their production is essentially not a deadly event of Gram-negative bacteria. Some Gram-positive bacteria alter the bacteriocin specific transport system, while others rely on sec-dependent export pathways [12].

Among Gram +ve bacteria, lactic acid bacteria are mainly used to produce bacteriocin. Bacteriocin has been categorized into three categories because of its quality, structure, and properties: Class I Bacteriocin, also known as lantibiotics, because they contain modified amino acids after translation such as lanthionine and beta-methyl lanolin [13]. Based on the structure and type of inhibition, it is further subdivided into subgroups A and B. Type A inhibits bacterial species by depolarizing the plasma membrane. They are usually in the range of 21 to 38 amino acids and bigger than the type B lantibiotics. Nisin is a type A lantibiotic who is best studied and commercially exploited bacteriocin. Type B lantibiotics inhibit bacteria by suppressing their enzymes e.g. mersacidin, it interferes with cell wall biosynthesis [14]

3.1. Bacteriocins produced by Streptococcus sp.

Most Streptococcus strains are testified to be pathogenic. Subsequently, bacteriocins isolated from Streptococcus are commonly pathogenic Streptococci [15]. The bacteriocins that have been characterized were initiated from few species i.e., *Streptococcus salivarius, S. pyogenes, S. macedonicus, S. mutans, S. bovis, S. uberis, S. thermophilus, S. rattus, S. phocae*. Most Streptococci bacteriocins were characterized to be lantibiotics among that cationic type A-lantibiotics are prevalent [16]. *S. salivarius* bacteriocin was the first Streptococci among lantibiotic characterized that efficiently inhibited human pathogen *Streptococcus pyogenes* [17].

Mutacine is a peptide bacteriocin produced by *Streptococcus mutans*. At least three different lantibiotics, namely the variant proteins I, II and III, have been reported from some *Streptococcus mutans* isolates. Mutacin II is structurally similar to the 481 groups of L-type lantibiotics [18]. It has been reported that *Streptococcus mutans* UA140 produces two peptide class II bacteriocins [Mutacin IV]. Nisin is most prominent lantibiotic bacteriocin that was reported from *Lactococcus lactis.* Recently, Nisin U produced by *Streptococcus uberis* revealed 78% identity to Nisin A, subsequently considered to be a Nisin variant [19].

A lantibiotic bovicin 255 produced by a *Streptococcus bovis* isolated from cow rumen, whereas a class II non-lantibiotic, bovicin HJ50 was reported from similar source by Xiao and co-workers [52]. In another study, two different peptide bacteriocins called BHT-A and BHT-B were reported from *Streptococcus rattus* [20]. During *S. pyogenes* screening, 10% of the strains were found to form bacteriocins. Consequently, a lantibiotic "Streptin" was produced by homogeneity that showed a molecular mass of 2.42 kDa [21].

Streptococcal bacteriocin in fermented foods has been reported. Macedocin is a lantibiotic from *S. macedonicus* that is isolated from handmade cheese [22]. Thermophilin 13, an IIb dipeptide bacteriocin, is produced by *Streptococcus thermophilus* isolated from yogurt. The antibiotic bacteriocin phocaecin PI80 is made from *S. phocae* PI80 isolated from the intestine of *Penaeus indicus*. Elimination or severe deterioration of the flora can lead to diarrhea or constipation, which is why it is necessary to maintain a healthy bacterial flora [23].

3.2. Bacteriocins produced by *Enterococcus faecium*

Enterococcus faecium is a gram-positive, homo-fermentative, lactic acid bacterium that is a natural inhabitant of the gastrointestinal tract. However, they are also found in fermented foods and are often separated from starter cultures and cheese producers. *E. faecium* bacteriocins have attracted attention in recent years because they are easily separated from several fermented foods and many of them remain active beside foodborne pathogens, such as *Listeria monocytogenes*. [24].

E. faecium T136 is sequestered as of Spanish dry fermented sausages producing enterocin A and B. They are effective against a variety of Gram-positive bacteria, including Listeria and Staphylococcus. Sequencing of N-terminal amino acids revealed the similarity of enterocin A with pediocin family of bacteriocins whereas enterocin B showed strong homology to carnobacteriocin A [25]. Enterobacterin P inhibits most of the food borne Gram +ve bacterial pathogens such as *Listeria monocytogenes, Clostridium perfringens, Staphylococcus aureus,* and *Clostridium botulinum*. It has been subjected to high temperature treatment [121°C for 15 minutes] and pH [2.0 ~ 11.0] for a wide range of effects, freezing, lyophilization, thawing, and long term storage at 4and 20°C [26].

Enterocin L50, originally called Pediocin L50, is a plasmid encoding broad spectrum bacteriocin produced by *Enterococcus faecium* L50. It is similar to a small group of cytolytic peptides secreted by certain staphylococci [27]. Enteromycin A, enteromycin B, and enterobacterin P-like bacteriocin have been reported from *Enterococcus faecium* JCM 5804. They inhibit the growth of *Clostridium*

botulinum, *Listeria monocytogenes* and vancomycin-resistant Enterococci [28].
In a similar manner, another research team reported anti-L. Monocyte proliferating bacteriocin-like inhibitors from *E. faecium* UQ31 [29]. Aymerich et al. describe the biochemical and genetic characteristics of Enteromycin A. [30] and reported that the Enterocin A leader sequence contains 18 amino acid residues. This is one of the other glycine precursors found in most other small non-lancholin bacteriocins, some lantibiotics and colicin V. Similarly, the gutcin I carried out was genetically characterized from E. 6T1a Enterococci and it was reported that the Enteromycin I was a bacteriocin of Pediocin family [31].

3.3. Bacteriocins produced by *Lactobacillus plantarum*

Lactobacillus plantarum can be isolated from different sources but mostly isolated from fermented food products. Todorov et al. [32] isolated *Lactobacillus plantarum* AMA-K from naturally fermented milk and characterized its bacteriocin. The presence of *Listeria inocula* stimulates the production of bacteriocins. *Lactobacillus plantarum* AMA-K grow in milk and produces only 800AU/ml in 24hrs. The bacteriocin ST8KF is isolated and characterized from kefir [33].
The temperature resistance of ST8KF up to 121°C for 15 to 20 minutes, indicated its bacteriostatic action. The bacterial pathogen *Lactobacillus plantarum* ST16Pa was first isolated by Todorov et al. [34] from papaya (*Carica papaya*). *Lactobacillus plantarum* ST16Pa is active against Listeria, Streptococcus, Pseudomonas, and Staphylococcus spp. In another study, *Lactobacillus plantarum* NCIM 2084 was identified to culture in glucose medium to yield bacteriocin. it's the bacteriocin produced by *Lactobacillus plantarum* NCIM 2084 is called as Planatricin LP84 which is bactericidal and cleavable counter to *E. coli* D21 and *Bacillus cereus* F4810 [35]. *Lactobacillus plantarum* ATCC8014 produced bacteriocin with 122kDa apparent molecular weight based on SDS-PAGE analysis can inhibit the growth of *E. coli*, *S. aureus*, *P. aeruginosa* and Lactobacillus resistant bacilli [36, 37]. The plant sterol ASM1 is classified as *L. plantarum* A-1. It is stable over a wide range of pH when it is heated it discharge a potential food bio-conservative. Xie et al., [38] reported an antibacterial pediocin LB-B1 from *L. plantarum* LB-B1 which was isolated from Chinese dairy products. The genome coding for pediocin LB-B1 was found 99.8% similar with the operon encoding pediocin PA-1, which indicate that both the bacteriocins are identical.

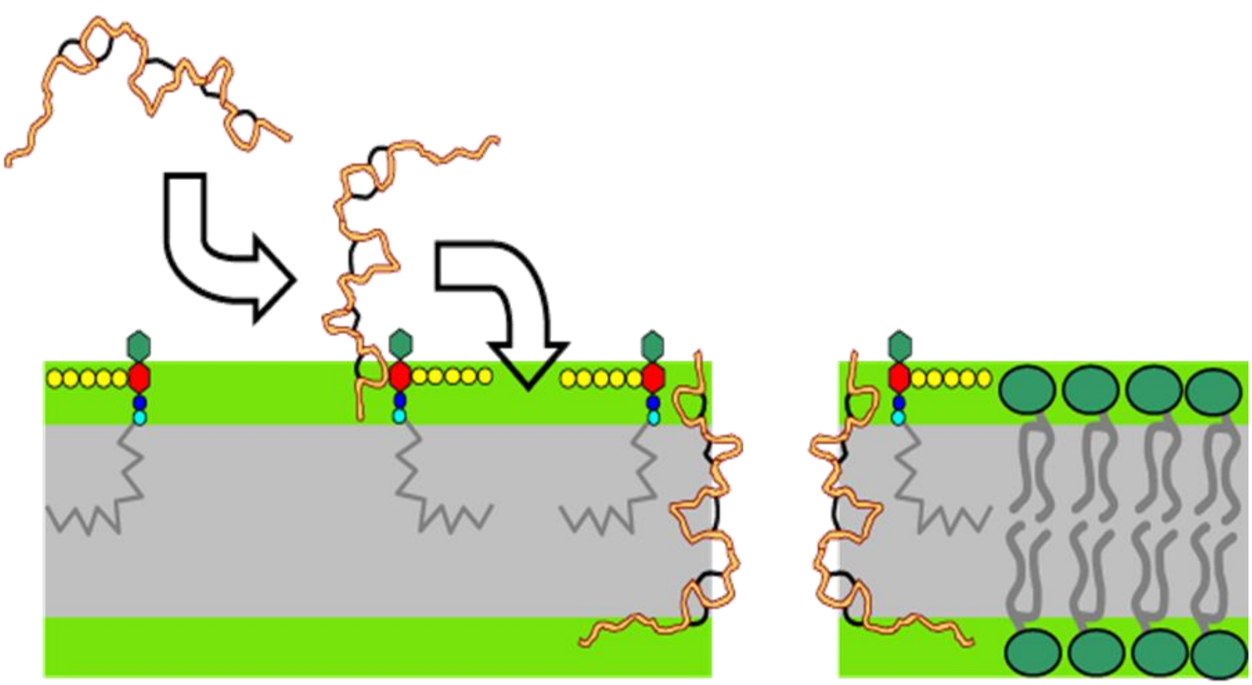

Figure 1: LAB produced bacteriocin mode of action [39]. Initially, Nisin interacts with Lipid II via N-terminus. Then Nisin induces the leakage of cytosolic contents by accumulating into the pore.

4. Probiotics in aquaculture

Bacteria are present in all corners of the water environment. In the life cycle of aquatic animals at the initial stage, they are unprotected to the attack of bacteria. Consequently, the presence of comparatively compact, non-pathogenic and different adherent microbiota on the egg may be an operative obstacle against the development of pathogenic colonies on the fish eggs. Additionally, the establishment of normal microbiota can be considered adjunct to the establishment of the digestive system and under normal conditions it serves as the barrier which inhibits the invasion of pathogens [40].

Larvae can take up a large number of bacteria. It is obvious that the microbiota of the egg influences the main colonization of larvae [41]. Kennedy et al. [42] used probiotics in the culture of marine fish larvae for ordinary snooks, red drums, spotted sea otters and striped fish. Then they believe that probiotics in larval aquaria [from egg through transformation] are used to increase survival, uniformity of size and growth rate. Regular inoculation of bacteria to the tank change the microbial community of the tank and fish.

Lara-Flores et al. [43] used two probiotics and *Saccharomyces cerevisiae* as growth promoters for *Nile tilapia* (*Oreochromis niloticus*). Their findings demonstrated that probiotic dietary supplement brooding has greater growth than brood feeding on a control diet. They also proposed that yeast is an additive to stimulate the growth in Tilapia farming. Gopalakannan & Arul, [44] isolated *Enterococcus faecium* MC13 from the gut of grey mullet *Mugil cephalus* and studied their protective effect on *Cyprinus carpio* after stimulating with pathogen *Aeromonas hydrophila*. In their study, a fish group treated with probiotic showed reduced mortality [22%] and fish were healthy but untreated fish group resulted in 100% mortality.

5. Fermented Foods as a source of LAB

Traditional fermented foods are widely used in prehistory and have become an important part of nutrition. They can be prepared at home or in the family craft with relatively simple techniques and equipments [45]. The fermentation was developed during the poverty phase as a preservation method and prevention technique against food spoilage. It is one of the oldest and most rational ways to produce and preserve food. In addition to preservation, fermented foods have the additional benefit of improving taste, increasing digestibility, and increasing nutritional value and pharmacological value [46].

LAB play an important role in the conservation and making of healthy fermented foods. Homologous and hetero-fermented LAB is often hard on synthetic media; on another hand, they tend to develop in most food matrices, rapidly lower the pH level so that other competing organisms can no longer grow. Leuconostoc and Nisin typically lower the pH to 4.0 to 4.5 and reduce some Lactobacilli and Sclerotia to about 3.5. Lactic acid bacteria contain most of the probiotic flora. Many LAB strains are generally considered safe (GRAS) and are commonly used in commercial foods for human consumption. Probiotics are single or mixed cultures of living microorganisms that favorably influence the host by enhancing the characteristics of the indigenous microbial community [47].

Lactic acid bacterial genera consist of Lactobacillus, Lactococcus, Enterococcus, Streptococcus, Pediococcus, Leuconostoc, Klebsiella spp. etc. In the Indian subcontinent, it is common to use fermented foods in local foods and other biological resources. However, the nature of products and basic materials varies region by region [48]. Fermented foods such as the development of Law and Daisy have been described in 700 BC by the Human Rights Commission. Today, there are hundreds of fermented foods with different raw materials and preparation methods. Each fermented food is associated with a unique group of micro-organisms that increase the levels of protein, vitamins, essential amino acids, and fatty acids. However, fermented foods have traditionally been made by spontaneous fermentation, and the knowledge of the microbiota of these products is limited.

5.1. Milk based fermented foods

Milk and milk-based products are widely consumed due to their nutritive value. For the same reason, milk is easily spoiled by pathogenic microorganisms; therefore, it is preferable to use lactic acid bacteria fermented milk for prevention. The lactic acid bacteria convert lactose-lactose into lactic acid and produce an antibacterial substance bacteriocin to inhibit spoilage bacteria. Dahi or quark is the most popular traditional Indian fermentation product produced by fermentation of lactic acid bacteria. Dahi or quark differs from yogurt as it uses a mixed starter made from *Lactococcus thermophilus*. The main taste of the induced metabolite is diacetyl, which is more valued by South Asians than the acetaldehyde flavor of yogurt [49].

Yak (*Bos grunnieus*, now called *Poephagus grunniens*) is one of the few domestic animals which can survive at extreme environmental conditions. It is normally found in the Highlands of Himalayas in Nepal, China [Tibet Plateau], India [Kashmir and Arunachal Pradesh], Bhutan and Mongolia. Milk is composed of 16.9-17.7g/L of dry matter, 55-72g/L of fat, 49-53g/l of proteins 8-9g/L of minerals and 45-50g/L lactose. There are a number of dairy products such as Chhur Churp, Chhu, cheese [Chhurpi], Shyow, Churkham, Maa and Philuk, and fermented milk (Kurut). The yak cheese is chemically composed of about 68.2% total solids (TS), 1.37% of salt and 49.4% dry matter [50]. It has been reported that LAB isolated from yogurt can be used to treat intestinal diseases such as diarrhea [51]. The species of LAB isolated from dairy products consist of *Streptococcus cremoris, S. thermophilus, S. lactis, Lactobacillus helveticus, L. Alimentarius, L. Hilgardii, L. Pseudoplantarum, L. acidophilus, L. bulgaricus, L. Kefir, L. Brevis, L. Farciminis, Leuconostoc mesenteroides, Lactococcus lactis subsp. cremoris, Enterococcus faecium, L. cremoris, L. bifermentans* and *L. casei subsp. Casei*.

5.2. Fruits and vegetable-based fermented foods

Lactic acid fermentation of vegetables, used as a method of preservation of finished and semi-finished products, is considered to be due to its ability to improve nutritional value, palatability, acceptance, quality, and durability of microbial plants, as a major technical fermentation product [53] In addition, this is a significant method for storing perishable vegetables with or without refrigeration, as the vast rural population does not have canned or frozen foods. Certain fermented vegetable products [gundruk, sinki, iniziangsang] are considered to be good appetizers, and ethnic people use these foods to treat indigestion [54]

Fermented bamboo shoots [BS] products are used as a traditional food by ethnic groups in northeast India [55]. In India, bamboo tin gold [26.2 tons], Meghalaya [435 tons] and Mizoram [426.8 tons] are harvested every year. Bamboo shoots contain a small amount of fat and cholesterol, but contain a lot of potassium, carbohydrates, and fiber. Many nutrient active materials (a.a and vitamins) and anti-oxidants (flavonoids, steroids, and phenols) can be extracted from bamboo shoots [56]. LAB is the major main microorganisms to prepare the fermented bamboo shoots and vegetables [57].

In fermented vegetables, *Pediococcus pentasaceous, Lactobacillus, Lactobacillus plantarum, Lactobacillus fermentum, Lactobacillus brevis, Enterobacter faecalis, Lactococcus lactis, Enterococcus faecium, and Lactobacillus acidophilus* are the LAB species. [58] identified the functional properties of LAB which were isolated from ethnically fermented vegetables in Himalayas (inziangsang, khalpi, gundruk, and sinki). LAB strains indicated strong acidification and coagulation activities. They showed antimicrobial activity, particularly a strain *L. plantarum* isolated from *inziangsang*, a fermented leafy vegetable product, was inhibitory towards *P. aeruginosa* and *S. aureus.*

LAB strains showed various enzymatic activities such as lipase, esterase, valine arylamidase, acid phosphatase, alkaline phosphatase, leucine arylamidase, cysteine-arylamidase, α-galactosidase, napthol-AS-B1-phosphohydrolase, β-galactosidase, β-glucosidase, α-glucosidase, N-acetyl-β-glucosaminidase and also degraded oligosaccharides. Some strains of *L. plantarum* showed more than 70% hydrophobicity and adherence to the mucus secreting HT-29 MTX cells. *L. plantarum* isolated from ayurvedic medicinal food *Kanji* or *Kanjika* is a potential source of Vitamin B12 [59]. During fermentation of radish tap root product, *sinki L. plantarum* utilizes mannitol to eliminate the bitter flavor from the finished product [60].

Bamboo shoot based fermented food products contain *L. plantarum, L. corniformis, L. brevis, L. fermentum, L. delbrueckii, Lactococcus lactis, Enterococcus durans*, based fermented foods contain *Lactobacillus plantarum, Streptococcus casei, L. lactis, Leuconostoc fallax, L. mesenteroides,* and *Tetragenococcus halophilus*, and as predominant LAB species, they also exhibited functional probiotic properties [61].

6. Conclusion

Worldwide, Lactic acid bacteria are usually allied with fermented dairy products, and the use of early dairy culture has become an industry in this century. In addition, lactic acid bacteria have been linked to health for many years. Today, many foods are particularly healthy due to the nature of some lactic acid bacteria. Unfortunately, all these items have not been carefully studied, so it may be difficult to confirm some claims. Therefore, it is clear that a critical study of the effect of stress selection and quality on health is necessary. Therefore, lactic acid bacteria and their products impart fermented food flavor, texture, and unique aroma while preventing spoilage and extending shelf life. Inhibition of the pathogenic microorganism by nisin a bactericidal protein can destroy Gram-negative bacteria under conditions that alter the exterior membrane.

Conflict of Interest

All authors have disclosed no conflicts of interest.

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